STUDIES ON THE ROLE OF *Drosophila buschii* IN THE DEVELOPMENT OF FRUIT ROT CAUSED BY *Aspergillus niger*

Part II

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A B S T R A C T

The studies deal with the interaction of Drosophila busckii, an insect and Aspergillus niger, a fungus causing fruit rot of tomato in storage and transit. D. busckii was very frequently encountered in vegetable shops and A. niger was a fungus commonly and frequently isolated from rotten fruits of tomato. Although thirteen fungi were found associated with fruit rot but the frequency of A. niger was high. A temperature 30°C and relative humidity of 95-100 percent were conducive for the development of fruit rot caused by A. niger both in the presence or absence of insect, D. busckii. The presence of the insect brought out early appearance and higher intensity of fruit rot. Thus probably the insect aggravated the fruit rot.

The fruit rot causing fungus was isolated from all parts of the insect body, indicating that the insect is able to transmit the fungus from one fruit to other and from one locality to other. The ascorbic acid content in the fruits of tomato decreased both in fungus inoculated fruits and insect fed but the decrease was more when the fruits were inoculated with fungus in the presence of insect. Similarly, the number of amino acids in the fruit was more in fruits infected with fungus and fed with insect either separately or together. Methionine was not detected in fruits inoculated with fungus in the presence of insect.

In an attempt to control the development of fruit rot caused by A. niger in the presence or absence of insect, extract of ninteen plant species were tried. Almost extract of all the plant species were effective in reducing the fruit rot in the presence or absence of insect to a varying degree. Extracts of Azadirachta indica, Lantana camara, Eucalyptus globulus, Allium sativum, A. cepa, Ocimum sanctum and Mentha arvensis were more effective.

Of the three preparations of leaf of A. indica viz., leaf powder, water and ethanolic extract of leaf tested, the water extract was found to be more effective in delaying the onset of fruit rot in the presence or absence of insect. Similarly, the water extract of other plants were more effective than ethanolic extract. Even ethanol itself was effective in delaying the onset but for short duration.

In amongst the flower and leaf extract of L. camara tested, the effectiveness of the latter was more in delaying the onset of fruit rot in the presence or absence of insect.

The latex of Calotropis procera and Euphorbia hirta also gave protection against these two to some extent but the former exhibited higher degree of effectiveness.

The fruits obtained from plants grown in soil amended with oil cakes remained free of infection with A. niger in the presence of insect for more days than those obtained from plants grown in unamended soil.

Most of the earlier studies deal either with infection with fungus or insect alone but the present studies show that the interaction between the two is not merely the insect acting as vector or providing avenues/wounds for the fungus to penetrate but also acting together in aggravating the fruit rot. Thus these studies in a way are advancement to our knowledge in understanding the two which form an interdisciplinary approach. The use of plant extracts for reducing the fruit rot save the environment from chemical pollution hazards. Thus fruits so treated with plant extract are fit for human consumption. Although plant extracts have been tried against insects and fungal pathogens separately but their exhibiting for effectiveness against both i.e., insect and fungus in the development of fruit rot, is a new approach.

When the extracts of different plants were incorporated in the media for rearing D. busckii, the development of flies from egg laying to emergence was adversely affected in almost all the plant extracts added to a varying degree. There was no egg laying and therefore, no further development in the media containing extracts of Lantana camara and Eucalyptus globulus. Moreover, the time required for egg laying and further developmental stages was more in the media with plant extracts than in the control.

The plant extracts can therefore, be used for controlling both fruit rot of tomato caused by A. niger and the insect, D. busckii.

CONTENTS

REVIEW OF LITERATURE

Insect pests of crops of economic importance have been controlled by pesticides for a long time. Various types of compounds of different nature have been tested from time to time and the toxicity levels have been worked out. The literature, although immense, has earlier been reviewed by Parrella et al., (1982), Knapp and Herald (1983), Yokoyama and Pritchard (1984) and Carroll (1984).

Metcalf and Flint (1973) have classified different control measures of insects into two categories (a) applied control measure (b) natural control measures. In the applied control measure insecticides have been used for controlling insect pests of crop plants in addition to other methods of control (David and Horsburgh, 1985, Rup and Chopra, 1985 and Chao and William, 1986). Although a successful method of control but continuous and intensive use of certain insecticides against various insect pests has resulted in the development of races or strains of insects sufficiently resistant to the action of the insecticides which necessitates a complete change in the control measure (Metcalf and Flint, 1973).

Moreover, all these chemicals are hazardous in nature and plants/products sprayed with insecticides become unworthy of consumption for a long time due to mammalian toxicity (Bindra et al., 1973). In order to overcome this drawback plant products have been used from time to time.

Amongst the insecticides of plant origin, nicotine sulphate from Nicotiana tabacum and N. rustica have long been tried. The use of nicotine was first made as a decoction of tobacco during 17th century for destroying soft bodied insects having sucking type of mouth parts. Later crude nicotine or nicotine sulphate was tested and was found satisfactory for insect control. Another important plant Pyrethrum sp. (Chrysanthemum spp.) has proved of great value. The insecticidal value of Pyrethrum was first discovered in Iran sometimes in 18th century. It has been successfully tried for large number of insects (Elliott et al., 1978). Large number of plants (68 species, of members of Leguminosae) have also been tried against several insects (Martin, 1975). A product rotenone has been isolated from such plants which is responsible for insect control (Arant, 1942). The most important plant used from the time immemorial is the parts of Azadirachta indica (commonly known as neem). Its leaves, berries and oils in

various forms have been reported for insect control. According to Ladd et al., (1984) prepupae and newly formed pupae were less susceptible but pupae three days old and older were not affected. When seed extract of A. indica was applied on beetle (Popillia japonica), there was increase in the duration of the immature stages. Length of prepupae and pupal stages were also increased by application of azadirachtin, a product from this plant. Similarly, Saxena et al., (1984) reported that neem (A. indica) cake significantly reduced the numbers of newly emerged females of Nilaparvata lugens when confined to plant treated with neem cake. Adult longevity, fecundity, oviposition and hatchability of eggs were, however, not adversely affected by neem cake application. Prabhaker et al., (1986) concluded that neem seed extract when incorporated in to an artificial diet at different concentration not only prolonged the development to adult flies but induced the mortality in all larval stages of Trichoplusia ni and Spodoptera exigua. In the lowest concentration only one reproductive T. ni adult female was produced. However, in S. exigua no pupae were formed regardless of the extract tested. Larval mortality of both the species obtained indicated an activity similar to that of other insect growth regulator. According to Rao and Srivastava (1984)

when larvae of Heliothis armigera were immersed for 5 min in a 1% concentration of pure neem kernel suspension or 1% suspension from water dispersible powder or 1% emulsion and were confined on treated sorghum, the gain in larval weight was much lower than on untreated sorghum, the larval mortality was high and percentage of larvae that pupated was very low. The pure kernel suspension was significantly more effective than the other formulations. Lakhani and Patel (1985) pointed out that neem seed powder, road, dust and ash could be used to protect stored bajri, Pennisetum typhoideum (P. americanum) against Ephestia cautella. Ash was the most effective and cheapest material in reducing damage to the bajri by about 9%. Moreover, egg hatch and adult emergence were low. However, neem seed powder failed to give satisfactory protection against insect.

Some chemicals obtained from plants when used in conjunction with pyrethroid, rotenoids, nicotine and certain organophosphorus and carbamate insecticides markedly increased the toxicity of the mixture. These include sesamin, sesamolin obtained from Sesamum indicum (Dudley and Bronson, 1944). Rotenones have been isolated from more than 60 plant species which are equally toxic to insects but less harmful to plants. The plants include species of

Tephrosia, Derris, Lonchocarpus, Millettia and Mundulea (Jones, 1942). Veratrum album and V. viride have been used as insecticide for over 100 years but due to high cost of production these were not widely recommended. Seeds of Schoenocaulon officinale, S. drummondii and S. texanum (f. Liliaceae) were also found to have insecticidal properties.

Woods of Picrasma excelsa and Quassia amara (f. Simarubaceae) available as chips and extracts) have been widely used for controlling wide range of insects. The roots and stem of Ryania speciosa (f. Flacourtiaceae) have been found to contain insecticidal components with ryanodine as the most important (McIndoo and Sievers, 1924). Mikolajczak et al., (1984) concluded that seed oils of many sapindaceous plants contain substantial amounts of cyanolipids. Several of these compounds stimulated the aggregation of the beetles Oryzaephilus surinamensis and Tribolium confusum. These cyanolipids were highly toxic and had antipupation effects on the insect. Binder and Waiss (1984) reported that extract of soyabean Glycine max resulted in larval deaths and failure to complete larval to pupal metamorphosis in Heliothis zea. Gossypol adversely affected larval and pupal weight, percentage of pupation and time to

pupae in H. virescens (Raulston et al., 1985). Similarly Chan and Tam (1985) reported the α-tomatine, a glyco-alkaloid present in solanaceous plants exert antibiotic effects on Ceratitis capitata larvae. Increased concentrations of α-tomatine resulted in decreased larval survival, lower pupal weights, an extended pupation period and a prolonged period of adult emergence. Pandey et al., (1979), while testing various plant extracts against insects reported that ether extracts of Acorus calamus rhizome gave highest percentage of larval mortality. Prabhu and John (1975) pointed out that extracts of certain plants possess juvenile hormone activity against nymphs of Dysdercus cingulatus. D. cingulatus laid normal eggs but the adults were found to have crumpled or small wings and three segmented tarsi.

In the present investigations, two organisms, one a fungus Aspergillus niger causing fruit rot of tomato in storage and an insect, Drosophila busckii the agent which help in dissemination of fungal spores have been studied. Both when present together, the fruit rot of tomato due to A. niger is aggravated. No fungicide or insecticide could be used because of different target organisms. Moreover, these chemicals if used on ripe tomatoes may render fruits

not worthy of consumption. Therefore, there is need to use plant products which are not only safe to use but harmless to consumers. In part I of the thesis it has been shown that the extract of certain plants when applied on fruits considerably reduced the incidence of fruit rot caused by A. niger both in presence/absence of D. busckii. Therefore, it became important to know the effect of the extracts of these plants on behaviour and development of the fruit fly, D. busckii.

MATERIALS AND METHODS

Rearing of *Drosophila busckii*

Flies were reared on banana-agar medium, containing 250 ml of crushed over ripe banana, 20g of yeast tablet and 20g of agar in 625 ml of distilled water. In order to prepare this medium, 20g of agar was mixed in boiling water until dissolved. To this was added 250 ml of crushed banana with 20g of yeast. This was mixed thoroughly. To avoid fungal growth a pinch of sodium benzoate was added. The medium was poured in the 150 ml of Erlenmyer flasks which were autoclaved for 15 min at 15 lb pressure.

Preparation of leaf extracts

Freshly collected and thoroughly washed leaves (25g) of different plants were ground in pestle and morter with 10 ml of distilled water and filtered through, Whatman filter paper No. 1 . The filtrate was named as 'standard' (S). The solution was sterilized by passing through Seitz filter and stored in refrigerator in sterile container.

Effect of various plant extracts on the egg laying, pupation and emergence of the flies

To each Erlenmyer flask containing 25 ml of agar medium was added leaf extract so sterilized in such quantity as to obtain 1% and 0.1% concentration.

Eggs of D. busckii (100) were released in each flask. The flasks were incubated at 28±1°C and checked daily until emergence of adult flies. The days required for larval emergence, pupation and adult flies were recorded. The number of larvae, pupae and adults were counted and percent emergence of fly was calculated.

The data were analysed by using analysis of variance.

Effect of leaf extracts of different plants on the behaviour of the flies

Tomato fruits dipped in different concentration of extract viz., standard, 1% and 0.1% various plants were kept in sterilized 250 ml beaker covered with muslin cloth.

The adults of Drosophila busckii reared in the above manner were released to see the behavioural changes in the flies. Changes in behaviour observed during their visit to treated fruits were noted after 5, 10 and 15 min intervals. Fruits treated with sterile distilled water were kept for control.

R E S U L T S

It is clear from Table 1 that normal egg laying took place in the medium containing standard extracts of Chenopodium album, Euphorbia hirta, Foeniculum vulgare, Ocimum sanctum, Peucedenum graveolans, Withania somnifera. However, in media with the remaining plant extracts there was no egg laying at all. Among those where egg laying took place, highest number of eggs was laid in the medium containing extracts of Solanum xanthocarpum and Foeniculum vulgare followed by E. hirta, W. somnifera and C. album. With increase in the dilution of the extract in the medium the number of eggs laid increased (Tables 2,3). In those where there was no egg laying in the media containing standard concentration of plant extract it was, however, observed after 6 days when 1% or 0.1% conc. of the extracts were incorporated except in Eucalyptus globulus and Lantana camara where there was no egg laying in any of the concentrations of plant extracts tested. In media containing plant extracts of different concentrations the egg laying was also delayed i.e., it occurred on 6th day in the media with different concentrations of plant extracts as against 2 days in control.

Table 1: Effect of (standard) extracts of different plants when incorporated in the medium on number of eggs laid by Drosophila busckii.

Plant species	S Number of eggs laid after(days)			
	2	4	6	8
Adenocalymna alliacea	-	-	-	-
Allium cepa	-	-	-	-
Allium sativum	-	-	-	-
Argemone mexicana	-	-	-	-
Azadirachta indica	-	-	-	-
Callistemon lanceolatus	-	-	-	-
Calotropis procera	-	-	-	-
Cassia fistula	-	-	-	-
Chenopodium album	-	-	10	15
Cymbopogon citratum	-	-	-	-
Eucalyptus globulus	-	-	-	-
Euphorbia hirta	-	-	15	19
Foeniculum vulgare	-	-	31	40
Lantana camara	-	-	-	-
Mentha arvensis	-	-	-	-
Ocimum sanctum	-	-	32	38
Peucedenum graveolans	-	-	-	-
Solanum xanthocarpum	-	-	31	38
Withania somnifera	-	-	12	18
Control	36	40	42	42

Table 2: Effect of (1% conc.) of extract of different plants when incorporated in the medium on number of eggs laid by Drosophila busckii

Plant species	1% Number of eggs laid after (days)			
	2	4	6	8
Adenocalymna alliacea	-	-	20	28
Allium cepa	-	-	20	30
Allium sativum	-	-	22	26
Argemone mexicana	-	-	31	36
Azadirachta indica	-	-	21	28
Callistemon lanceolatus	-	-	31	37
Calotropis procera	-	-	22	27
Cassia fistula	-	-	33	40
Chenopodium album	-	-	20	30
Cymbopogon citratum	-	-	22	26
Eucalyptus globulus	-	-	-	-
Euphorbia hirta	-	-	22	28
Foeniculum vulgare	-	-	40	40
Lantana camara	-	-	-	-
Mentha arvensis	-	-	21	29
Ocimum sanctum	-	-	40	40
Peucedenum graveolans	-	-	21	28
Solanum xanthocarpum	-	-	40	40
Withania somnifera	-	-	22	25
Control	36	40	42	42

Table 3: Effect of (0.1% Conc.) of extracts of different plants when incorporated in the medium on number of eggs laid by Drosophila busckii

Plant species	0.1% Number of eggs laid after (days)			
	2	4	6	8
Adenocalymna alliacea	-	-	31	34
Allium cepa	-	-	34	38
Allium sativum	-	-	32	39
Argemone mexicana	-	-	40	40
Azadirachta indica	-	-	18	20
Callistemon lanceolatus	-	-	40	40
Calotropis procera	-	-	35	40
Cassia fistula	-	-	40	40
Chenopodium album	-	-	33	37
Cymbopogon citratum	-	-	31	38
Eucalyptus globulus	-	-	-	-
Euphorbia hirta	-	-	32	38
Foeniculum vulgare	-	-	40	40
Lantana camara	-	-	-	-
Mentha arvensis	-	-	32	36
Ocimum sanctum	-	-	40	40
Peucedenum graveolans	-	-	32	37
Solanum xanthocarpum	-	-	40	40
Withania somnifera	-	-	33	40
Control	36	40	42	42

Table 3a: (Summary table). Effect of different concentrations of extracts of plants when incorporated in the medium on number of eggs laid by *Drosophila busckii* after 8 days.

Plant species	Number of eggs laid after 8 days in (conc.)		
	Standard	1%	0.1%
Adenocalymna alliacea	-	28	34
Allium cepa	-	30	38
Allium sativum	-	26	39
Argemone mexicana	-	36	40
Azadirachta indica	-	28	20
Callistemon lanceolatus	-	37	40
Calotropis procera	-	27	40
Cassia fistula	-	40	40
Chenopodium album	15	30	37
Cymbopogon citratum	-	26	38
Eucalyptus globulus	-	-	-
Euphorbia hirta	19	28	38
Foeniculum vulgare	40	40	40
Lantana camara	-	-	-
Mentha arvensis	-	29	36
Ocimum sanctum	38	40	40
Pencedenum graveolans	-	28	37
Solanum xanthocarpum	38	40	40
Withania somnifera	18	25	40
Control	42	42	42
L.S.D. 5%	4.3	5.3	4.8
1%	6.5	7.2	5.5

There was no hatching of larvae in the media containing 'S' concentration of Adenocalymna alliacea, Allium cepa, A. sativum, Argemone mexicana, Azadirachta indica, Callistemon lanceolatus, Calotropis procera, Cassia fistula, Cymbopogon citratum, E. globulus, L. camara, M. arvensis and P. graveolans. Hatching initiated after 4 days and it was 60% in F. vulgare. In O. sanctum and S. xanthocarpum it initiated after 6 days and in W. somnifera and Euphorbia hirta after 8 days whereas in control it occurred within 2 days. Of the various plant extracts where hatching occurred, maximum hatching was observed in the presence of extracts of F. vulgare and O. sanctum after 10 days though they differed in the initiation of hatching; (in the latter it was delayed). However, the hatching was least in E. hirta (Table 4). When the concentration of plant extract in the medium was 1%, hatching occurred in all the plant extracts containing media except Allium cepa, Azadirachta indica, Eucalyptus globulus, Lantana camara and Mentha arvensis. Maximum hatching took place in the media containing 1% extract of Foeniculum vulgare, Ocimum sanctum and least in the media containing Adenocalymna alliacea. The onset of hatching or time required for hatching ranged from 4-8 days as against 2 days in the media containing Allium sativum, Chenopodium album,

Table 4: Effect of (Standard) extracts of different plants when incorporated in the medium on hatching of *Drosophila busckii*

Plant species	No. of larvae hatched after (days)				
	2	4	6	8	10
Adenocalymna alliacea	-	-	-	-	-
Allium cepa	-	-	-	-	-
Allium sativum	-	-	-	-	-
Argemone mexicana	-	-	-	-	-
Azadirachta indica	-	-	-	-	-
Callistemon lanceolatus	-	-	-	-	-
Calotropis procera	-	-	-	-	-
Cassia fistula	-	-	-	-	-
Chenopodium album	-	-	10	15	15
Cymbopogon citratum	-	-	-	-	-
Eucalyptus globulus	-	-	-	-	-
Euphorbia hirta	-	-	-	8	8
Foeniculum vulgare	-	60	60	65	65
Lantana camara	-	-	-	-	-
Mentha arvensis	-	-	-	-	-
Ocimum sanctum	-	-	68	70	70
Peucedenum graveolans	-	-	-	-	-
Solanum xanthocarpum	-	-	45	46	46
Withania somnifera	-	-	-	10	15
Control	80	80	81	81	81

Euphorbia hirta, Foeniculum vulgare, Ocimum sanctum, Solanum xanthocarpum; 6 days in Adenocalymna alliacea, Cassia fistula, Cymbopogon citratum and Withania somnifera; and 8 days in Argemone mexicana and Peucedenum graveolans. With further dilution of the extract in the medium to 0.1%, hatching occurred even in some of those plant extracts containing media where in 1% conc containing media it did not occur. After 10 days hatching occurred in all the plant extracts except E. globulus and L. camara. The onset of hatching ranged from 4-6 days. The time required for the initiation of hatching was 4 days in the media containing 0.1% conc. extract of Allium sativum, Argemone mexicana, Azadirachta indica, Cassia fistula, Chenopodium album, Cymbopogon citratum, Euphorbia hirta, Foeniculum vulgare, Ocimum sanctum, Peucedenum graveolans, Solanum xanthocarpum and Withania somnifera and 6 days in the media containing 0.1% extract of Adenocalymna alliacea, Allium cepa, Callistemon lanceolatus, Calotropis procera and Mentha arvensis. Highest hatching occurred in O. sanctum, Chenopodium album, F. vulgare and S. xanthocarpum and least in Azadirachta indica. Even in this concentration of plant extract containing media there was no hatching when extract of E. globulus and L. camara were added to the media (Tables 5,6).

Table 5: Effect of (1% Conc.) of extracts of different plants when incorporated in the medium on hatching of *Drosophila busckii*

Plant species	No. of larvae hatched after (days)				
	2	4	6	8	10
Adenocalymna alliacea	-	-	8	10	10
Allium cepa	-	-	-	-	-
Allium sativum	-	60	60	60	60
Argemone mexicana	-	-	-	12	15
Azadirachta indica	-	-	-	-	-
Callistemon lanceolatus	-	-	-	15	18
Calotropis procera	-	-	-	10	15
Cassia fistula	-	-	12	15	20
Chenopodium album	-	60	68	68	68
Cymbopogon citratum	-	-	15	18	18
Eucalyptus globulus	-	-	-	-	-
Euphorbia hirta	-	8	15	35	45
Foeniculum vulgare	-	68	68	70	70
Lantana camara	-	-	-	-	-
Mentha arvensis	-	-	-	-	-
Ocimum sanctum	-	64	68	70	70
Peucedenum graveolans	-	-	-	18	35
Solanum xanthocarpum	-	40	55	58	68
Withania somnifera	-	-	25	35	45
Control	80	80	81	81	81

Table 6: Effect of (0.1% Conc.) of extracts of different plants when incorporated in the medium on hatching of *Drosophila busckii*

Plant species	No. of larvae hatched after (days) 2	4	6	8	10
Adenocalymna alliacea	-	-	30	30	30
Allium cepa	-	-	34	38	38
Allium sativum	-	60	60	64	64
Argemone mexicana	-	15	25	28	28
Azadirachta indica	-	8	8	10	16
Callistemon lanceolatus	-	-	18	28	28
Calotropis procera	-	-	30	35	36
Cassia fistula	-	18	40	40	40
Chenopodium album	-	68	68	68	68
Cymbopogon citratum	-	18	38	48	48
Eucalyptus globulus	-	-	-	-	-
Euphorbia hirta	-	18	38	48	48
Foeniculum vulgare	-	68	68	68	68
Lantana camara	-	-	-	-	-
Mentha arvensis	-	-	50	55	58
Ocimum sanctum	-	70	70	74	74
Peucedenum graveolans	-	18	38	38	38
Solanum xanthocarpum	-	51	68	68	68
Withania somnifera	-	26	38	58	58
Control	80	80	81	81	81

Table 6a (Summary table): Effect of different concentrations of extracts of plants when incorporated in the medium on number of larvae hatched after 8 days.

Plant species	Number of larvae hatched after 8 days in (conc.) Standard	1%	0.1%
Adenocalymna alliacea	-	10	30
Allium cepa	-	-	38
Allium sativum	-	60	64
Argemone mexicana	-	12	28
Azadirachta indica	-	-	10
Callistemon lanceolatus	-	15	28
Calotropis procera	-	10	35
Cassia fistula	-	15	40
Chenopodium album	15	68	68
Cymbopogon citratum	-	18	48
Eucalyptus globulus	-	-	-
Euphorbia hirta	8	35	48
Foeniculum vulgare	65	70	68
Lantana camara	-	-	-
Mentha arvensis	-	-	55
Ocimum sanctum	70	70	74
Peucedenum graveolans	-	18	38
Solanum xanthocarpum	46	58	68
Withania somnifera	10	35	58
Control	81	81	81
L.S.D. 5%	5.8	7.9	6.3
1%	8.3	9.8	9.4

No pupation took place in the media containing extracts of L. camara and E. globulus in any of the three concentrations tested. In the media containing extracts of Allium cepa and M. arvensis the pupation took place after 4 days only when the concentration of the extract in the media was 0.1%. Of the two species Allium cepa appeared to be more effective as the number of developing pupae was less than M. arvensis. In the media containing extracts of Allium sativum and Azadirachta indica the pupation took place after 4 days when the concentration of the extract was 1% and 0.1%. The number of pupae developing in the medium containing Azadirachta indica was less as compared to the medium containing Allium sativum. In the media with remaining plant extracts the pupation occurred in all the three concentrations tested. The number of pupae emerged was maximum in the media with 0.1% concentration of Chenopodium album and lowest in A. indica (Table 7).

By and large Azadirachta indica appeared to be more effective in amongst those where pupation occurred than any other plant extracts. Although pupation took place in the media with 1% and 0.1% but the number of pupae emerged was low throughout.

Table 7: Effect of different concentrations of extracts of plants when incorporated in the medium on pupation in *Drosophila busckii*

Plant species	No. of pupae in different concentration after (days)											
	2	4	6	8	2	4	6	8	2	4	6	8
Adenocalymna alliacea	-	-	-	-	-	20	20	20	-	25	25	25
Allium cepa	-	-	-	-	-	-	-	-	-	28	28	28
Allium sativum	-	-	-	-	-	51	51	51	-	55	55	55
Argemone mexicana	-	29	29	29	-	30	30	30	-	32	32	32
Azadirachta indica	-	-	-	-	-	3	3	3	-	3	3	3
Callistemon lanceolatus	-	35	35	35	-	38	38	38	-	40	40	40
Calotropis procera	-	22	22	22	-	24	24	24	-	28	28	28
Cassia fistula	-	30	30	30	-	35	35	35	-	38	38	38
Chenopodium album	-	65	65	65	-	68	68	68	-	71	71	71
Cymbopogon citratum	-	32	32	32	-	35	35	35	-	37	37	37
Eucalyptus globulus	-	-	-	-	-	-	-	-	-	-	-	-
Euphorbia hirta	-	38	38	38	-	42	42	42	-	50	50	50
Foeniculum vulgare	-	60	60	60	-	64	64	64	-	66	66	66
Lantana camara	-	-	-	-	-	-	-	-	-	-	-	-
Mentha arvensis	-	-	-	-	-	-	-	-	-	42	42	42
Ocimum sanctum	-	60	60	60	-	60	60	60	-	60	60	60
Peucedenum graveolans	-	28	28	28	-	30	30	30	-	32	32	32
Solanum xanthocarpum	-	62	62	62	-	65	65	65	-	69	69	69
Withania somnifera	-	58	58	58	-	60	60	60	-	63	63	63
Control	-	80	80	80	-	80	80	80	-	80	80	80

Table 7a (Summary table): Effect of different concentrations of extracts of plants when incorporated in the medium on number of pupae emerged after 8 days.

Plant species	Number of pupae emerged after 8 days in (Conc.) Standard	1%	0.1%
Adenocalymna alliacea	-	20	25
Allium cepa	-	-	28
Allium sativum	-	51	55
Argemone mexicana	29	30	32
Azadirachta indica	-	3	3
Callistemon lanceolatus	35	38	40
Calotropis procera	22	24	28
Cassia fistula	30	35	38
Chenopodium album	65	68	71
Cymbopogon citratum	32	35	37
Eucalyptus globulus	-	-	-
Euphorbia hirta	38	42	50
Foeniculum vulgare	60	64	66
Lantana camara	-	-	-
Mentha arvensis	-	-	42
Ocimum sanctum	60	60	60
Peucedenum graveolans	28	30	32
Solanum xanthocarpum	62	65	69
Withania somnifera	58	60	63
Control	80	80	80
L.S.D. 5%	8.5	7.8	8.5
1%	10.9	9.5	10.3

In the media containing extracts of E. globulus and L. camara no adult emerged in any of the three concentrations tested even after 10 days of incubation. Adenocalymna alliacea, Allium sativum and Azadirachta indica did not favour emergence of adults in the standard concentration. But flies emerged in the media with 1% of all the different plant extracts except L. camara, E. globulus and M. arvensis and least number of adults emerged in the medium having the extract of A. indica. In the medium with 0.1% conc. maximum number of adults emerged in Chenopodium album and S. xanthocarpum and least in Azadirachta indica. This was true with the media containing other two concentrations of the plant extracts where adults emerged. It, therefore, appears that extracts of L. camara, E. globulus did not support egg laying, hatching, pupation and emergence of flies. Amongst those where development of the fly was observed extracts of A. indica was most effective as figures of egg laying and hatching were below 50/100 eggs. Further development was arrested as number of pupae formed was three and only two flies emerged. (Table 8)

When fruits were treated with different concentrations of plant extracts and flies were released over them in a beaker, the behaviour was not normal. Over the fruit,

Table 8: Effect of different concentrations of extracts of plants when incorporated in the medium on adults of *Drosophila busckii*

Plant species	No. of adults in different concentration after (days)											
	2	4	6	8	2	4	6	8	2	4	6	8
Adenocalymna alliacea	-	-	-	-	-	15	15	15	-	18	18	18
Allium cepa	-	20	20	20	-	20	20	20	-	20	20	20
Allium sativum	-	-	-	-	-	46	46	46	-	50	50	50
Argemone mexicana	-	22	22	22	-	25	25	25	-	28	28	28
Azadirachta indica	-	-	-	-	-	2	2	2	-	2	2	2
Callistemon lanceolatus	-	31	31	31	-	32	32	32	-	33	33	33
Calotropis procera	-	21	21	21	-	25	25	25	-	26	26	26
Cassia fistula	-	25	25	25	-	28	28	28	-	30	30	30
Chenopodium album	-	61	61	61	-	63	63	63	-	66	66	66
Cymbopogon citratum	-	28	28	28	-	30	30	30	-	32	32	32
Eucalyptus globulus	-	-	-	-	-	-	-	-	-	-	-	-
Euphorbia hirta	-	35	35	35	-	41	41	41	-	45	45	45
Foeniculum vulgare	-	55	55	55	-	58	58	58	-	62	62	62
Lantana camara	-	-	-	-	-	-	-	-	-	-	-	-
Mentha arvensis	-	-	-	-	-	-	-	-	-	38	38	38
Ocimum sanctum	-	50	50	50	-	52	52	52	-	52	52	52
Peucedenum graveolans	-	23	23	23	-	25	25	25	-	26	26	26
Solanum xanthocarpum	-	50	50	50	-	55	55	55	-	60	60	60
Withania somnifera	-	52	52	52	-	54	54	54	-	58	58	58
Control	-	78	78	78	-	78	78	78	-	78	78	78

Table 8a (Summary table): Effect of different concentrations of extracts of plants when incorporated in the medium on number of adults emerged after 8 days.

Plant species	Number of adults emerged after 8 days in (Conc.) Standard	1%	0.1%
Adenocalymna alliacea	-	15	18
Allium cepa	20	20	20
Allium sativum	-	46	50
Argemone mexicana	22	25	28
Azadirachta indica	-	2	2
Callistemon lanceolatus	31	32	33
Calotropis procera	21	25	26
Cassia fistula	25	28	30
Chenopodium album	61	63	66
Cymbopogon citratum	28	30	32
Eucalyptus globulus	-	-	-
Euphorbia hirta	35	41	45
Foeniculum vulgare	55	58	62
Lantana camara	-	-	-
Mentha arvensis	-	-	38
Ocimum sanctum	50	52	52
Peucedenum graveolans	23	25	26
Solanum xanthocarpum	50	55	60
Withania somnifera	52	54	58
Control	78	78	78
L.S.D. 5%	10.5	9.8	8.3
1%	13.8	12.1	10.5

treated with standard extracts of plants emitting smell such as Eucalyptus globulus, Mentha arvensis, Ocimum sanctum, Allium cepa, A. sativum, Adenocalymna alliacea, Azadirachta indica, Callistemon lanceolatus and Cymbopogon citratum the flies were repelled at the very outset. The repelling period however, varied with the extract. In E. globulus it was 8-10 min in O. sanctum, 5-7 min, in A. indica, Allium cepa, A. sativum 5-7 min, in A. indica, Allium cepa, A. sativum, Callistemon lanceolatus, Cymbopogon citratum, 2-5 min. Later they adhered to the walls of the beaker without indication of movement. In the extracts emitting strong smell such as A. sativum and A. cepa they become unconscious for a period ranging from 5-10 min. (Table 10)

After regaining consciousness or when the repelling period was over, flies started moving brushing their legs and wings starting from upper side of the fruit to the lower side. Thus flies appear to be very sensitive to the pungent smell. In others, the flies tried to sit on the fruit so treated without success for certain period. When the water extract of plants dried up the sitting period and frequency of sitting of flies increased.

Table 9: LD50 of extracts of different plants for adults of *Drosophila busckii*

Plant species	%
Adenocalymna alliacea	> 0.1
Allium cepa	> 0.1
Allium sativum	> 0.1
Argemone mexicana	> 0.1
Azadirachta indica	> 0.01
Callistemon lanceolatus	> 0.1
Calotropis procera	> 0.1
Cassia fistula	> 0.1
Chenopodium album	10
Cymbopogon citratum	> 0.1
Eucalyptus globulus	-*
Euphorbia hirta	> 0.1
Foeniculum vulgare	10
Lantana camara	-
Mentha arvensis	> 0.1
Ocimum sanctum	10
Peucedenum graveolans	> 0.1
Solanum xanthocarpum	10
Withania somnifera	10

*Either there was no emergence of flies or if emerged the percent emergence was below 50%.

Table 10: Effect of plant extracts (standard conc.) on the behaviour of flies of *Drosophila busckii*

Plant species	Hovering period (min)	Repellent period (min)	Movement duration (min)				Movement period on fruits (min.)			Duration on fruit (min)	Frequency of visit fruits per mi
			Wings	Legs	ant portion	Abdomen	tip	Middle	base		
Adenocalymna alliacea	-	5	3	2	-	4	-	5	7	9	2
Allium cepa	-	2	5	-	-	5	2	5	-	8	3
Allium sativum	-	5	3	-	3	-	2	2	-	7	2
Argemone mexicana	-	2	2	3	-	-	2	2	5	8	3
Azadirachta indica	-	5	-	2	2	-	-	2	-	4	2
Callistemon lanceolatus	-	2	2	2	-	2	2	5	2	7	4
Calotropis procera	-	2	1	4	2	-	2	4	5	6	4
Cassia fistula	-	-	3	2	-	2	3	3	5	10	3
Chenopodium album	2	-	2	3	1	2	3	3	2	10	4
Cymbopogon citratum	-	5	2	-	2	2	2	2	2	8	5
Eucalyptus globulus	-	10	-	-	2	2	1	2	-	3	2
Euphorbia hirta	-	2	1	3	-	2	-	1	2	6	4
Foeniculum vulgare	-	-	2	2	1	2	1	3	3	10	4
Lantana camara	-	5	-	2	-	-	2	1	-	5	2
Mentha arvensis	-	2	-	3	-	3	2	3	4	10	3
Ocimum sanctum	-	5	2	2	-	2	-	2	2	7	3
Peucedenum graveolans	-	-	2	2	1	2	3	6	8	12	4
Solanum xanthocarpum	-	2	2	3	2	2	2	3	3	10	4
Withania somnifera	-	-	1	2	-	2	2	5	5	10	3

With increase in the dilution of the extract the period of abnormal behaviour decreased. In most of the cases the flies sat on the fruits but for shorter duration. The sitting period was hardly 2-4 minutes over the fruits treated with Lantana camara, Azadirachta indica, Allium cepa, A. sativum, Eucalyptus globulus, O. sanctum and Mentha arvensis but in the remaining it was for long duration. (Tables 11, 12)

Table 11: Effect of plant extracts (1%) on the behaviour of flies of *Drosophila busckii*

Plant species	Hovering period (min)	Repellent period (min)	Movement duration (min)				Movement period on fruits (min)			Duration on fruit (min)	Frequency of visit to fruit per min.
			Wings	Legs	ant portion	Abdomen	tip	middle	base		
Adenocalymna alliacea	2	2	3	2	2	4	2	5	7	12	3
Allium cepa	3	2	3	3	-	2	2	5	4	10	4
Allium sativum	3	2	4	2	1	2	2	3	2	8	3
Argemone mexicana	4	1	3	5	-	-	3	3	4	9	3
Azadirachta indica	2	4	-	2	-	2	1	2	-	6	3
Callistemon lanceolatus	4	2	2	2	3	-	3	5	2	9	5
Calotropis procera	3	2	3	1	2	3	3	4	5	8	5
Cassia fistula	3	1	2	2	2	2	5	3	5	10	4
Chenopodium album	4	2	2	3	2	1	4	3	3	12	5
Cymbopogon citratum	4	2	1	3	1	2	3	3	4	10	6
Eucalyptus globulus	-	8	2	2	-	-	1	2	1	5	3
Euphorbia hirta	2	1	3	3	2	2	2	1	2	8	5
Foeniculum vulgare	5	-	2	2	1	2	2	3	3	12	5
Lantana camara	2	3	1	2	-	-	2	1	2	7	3
Mentha arvensis	2	1	2	3	-	1	2	4	4	11	4
Ocimum sanctum	3	3	2	1	2	1	2	3	3	9	5
Peucedenum graveolans	3	-	3	3	2	2	4	5	5	10	5
Solanum xanthocarpum	3	1	2	4	2	4	3	4	5	8	5
Withania somnifera	4	-	2	2	3	2	3	4	4	8	6

Table 12: Effect of plant extracts (0.1%) on the behaviour of flies of *Drosophila busckii*

Plant species	Hovering period (min)	Repellent period (min)	Movement duration (min)				Movement period on fruits (min.)			Duration on fruits (min)	Frequency of visit t fruit per min.
			Wings	Legs	ant portion	Abdomen	tip	middle	base		
Adenocalymna alliacea	3	1	4	2	2	4	3	5	7	12	3
Allium cepa	4	1	3	3	1	2	4	5	4	10	4
Allium sativum	4	1	4	2	1	3	4	4	3	9	3
Argemone mexicana	5	-	3	5	1	-	3	4	6	10	4
Azadirachta indica	2	2	1	2	-	2	2	2	1	7	3
Callistemon lanceolatus	5	-	3	3	2	-	5	5	3	10	5
Calotropis procera	4	-	3	2	2	3	3	5	5	9	5
Cassia fistula	4	-	3	3	2	2	5	5	6	10	6
Chenopodium album	5	-	3	3	2	2	5	6	7	12	5
Cymbopogon citratum	5	-	2	3	1	2	3	5	6	12	6
Eucalyptus globulus	1	5	2	2	-	1	1	3	1	7	3
Euphorbia hirta	3	-	3	4	2	2	3	5	7	10	5
Foeniculum vulgare	5	-	3	3	1	3	3	4	5	12	5
Lantana camara	2	2	2	3	-	1	3	4	5	9	3
Mentha arvensis	4	-	3	3	-	2	5	5	5	12	4
Ocimum sanctum	4	2	3	3	-	2	4	5	5	11	5
Peucedenum graveolans	5	-	4	3	-	3	5	7	8	10	5
Solanum xanthocarpum	3	-	3	4	3	2	3	5	6	10	5
Withania somnifera	5	-	3	3	-	4	3	6	7	12	6

DISCUSSION

In earlier part of these studies it was shown that extracts of certain plants were able to reduce the incidence of fruit rot of tomato caused by Aspergillus niger both in the presence and absence of the insect, Drosophila busckii. In view of this it was considered desirable to determine the effect of these plant extracts on development of the insect. Out of the extracts of nineteen plant species tested in 'standard' extract highest egg laying was observed in the medium containing extract of Solanum xanthocarpum and Foeniculum vulgare followed by Euphorbia hirta, Withania somnifera and Chenopodium album. The egg laying improved in the dilution of extract. There has been no egg laying in the medium with Eucalyptus globulus and Lantana camara in any of the three concentrations tested. In the media with those plant extracts where egg laying occurred, it has been very poor in the medium with 1% and 0.1% concentrations of the extract of Azadirachta indica. The oviposition was also delayed in the media containing plant extracts. The egg laying in the media with different concentrations of plant extracts was observed after 4-8 hrs depending upon the type

of plant extracts added as against 2 hrs in control. Thus plant extracts appeared to have influenced the number of eggs laid and the initiation of the oviposition. Plant extracts have been reported to contain compounds like alkaloids, flavanoids, phenolic substances already known for toxicity to various kinds of organisms (Bhakuni et al., 1969, Smith, 1976 and Giannasi, 1978). It is likely that these might be affecting the oviposition in the present studies.

Emergence of larvae has not been observed in the media containing standard extract of Adenocalymna alliacea, Allium cepa, A. sativum, Argemone mexicana, Azadirachta indica, Callistemon lanceolatus, Calotropis procera, Cassia fistula, Cymbopogon citratum, Eucalyptus globulus, Lantana camara, Mentha arvensis and Peucedenum graveolans but media containing dilutions of 1% and 0.1% plant extract, hatching has been observed in all the above except E. globulus and L. camara. The emergence of larvae has been very poor in the extracts of A. indica. The larval emergence has been observed only after 6 days in A. alliacea, Allium cepa, Callistemon lanceolatus, Calotropis procera and Mentha arvensis. However, in the remaining where emergence occurred the onset has been after 4 days as against

2 days in control. Similar results were observed with emergence of pupae and adults. Leuschner (1972) also noted disruption of normal growth and development of nymphs of East African coffee bug, Antestiopsis orbitalis bechuana by the extracts of Azadirachta indica. This is in conformity with the results of the present studies where poor development of flies was observed. Nakanishi (1975) also postulated that azadirachtin a product from A. indica is similar to ecdysteroids or inhibitor of ecdysis. Probably this might be one of the factors in poor development of the insect. Besides azadirachtin, thionimone and nimbidin are also constituents of Azadirachta indica which might be affecting the normal development of flies.

No hatching of larvae and no emergence of flies in E. globulus and L. camara plant extract could be due to presence of highly toxic compounds in these plant extracts. L. camara is already reported as highly toxic to insects. (Prabhu and John, 1975 and Pandey et al., 1979). E. globulus is already known for certain aromatic compounds and essential oils which are antimicrobial and insecticidal (Barde, 1985). It is likely that these factors might be operating in the present studies in exhibiting the toxicity. The oil of Cymbopogon citratum has already been found to have the insect repellent property and have been commercialised as

mosquito repellents (Metcalf and Flint, 1973). In the present studies Cymbopogon citratum extract has been affective against oviposition and hatching in standard concentration only but in diluted concentration it is likely that some of the volatile substances are lost and its efficacy is reduced. Although, extracts of L. camara and E. globulus have been found very effective against the development of flies, i.e., no oviposition and no adult formation, but there are some plants where oviposition takes place but no emergence of larvae and no further development has been observed. This difference might be due to different compounds with different properties present in the extracts. LD_{50} values also indicate the least effectiveness of extracts of Chenopodium album, Foeniculum vulgare, Ocimum sanctum, Solanum xanthocarpum and Withania somnifera as more than 10 percent concentration in the medium is required for 50% effectiveness. (Table 9)

It appears that plant extracts of E. globulus and L. camara could be successfully used for controlling insect visits to fruits. In the extracts of plants found adversely affecting the development of insect, the behaviour of insect has been slightly different than the normal. At the very outset they were repelled and became unconscious for sometime.

It appears that the volatile products affect the sensory organs and nervous system in the insect (Anonymous, 1969) which varies with the type of volatile substances present. Therefore, some repell for longer and other for short duration. It is interesting to note that extracts of plants which proved very effective against fruit rot of tomato caused by A. niger are also found effective against insect development.

To summarise it can be suggested that extracts of certain plants are very effective against insect as well as fungus causing fruit rot (A. niger) and could be used for controlling the disease, and insect menace. There is another advantage that the application of extracts does not create any environmental pollution hazards.

SUMMARY

Studies were made to determine the effect of extracts of nineteen plant species when incorporated in the medium (banana-agar) on egg laying, larval hatching, pupation and development of adults of Drosophila busckii. There was complete inhibition of egg laying and further development in the medium containing extracts of Lantana camara and Eucalyptus globulus.

Amongst those plant extracts where egg laying and further development of flies took place when incorporated in the medium, highest reduction was found in the media containing extracts of Azadirachta indica.

Increase in dilution of the extracts incorporated in the media resulted in decreased efficacy of the extracts in inhibiting egg laying and further development.

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STUDIES ON THE ROLE OF *Drosophila buschii* IN THE DEVELOPMENT OF FRUIT ROT CAUSED BY *Aspergillus niger*

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This is to certify that the thesis entitled " Studies on the role of Drosophila busckii in the development of fruit rot caused by Aspergillus niger " submitted by Miss Purnima Sinha for the award of degree of Doctor of Philosophy of the Aligarh Muslim University, Aligarh is her own work carried out in our supervision and guidance. The work of inter-disciplinary nature was carried out both in the Departments of Botany and Zoology and is fit for submission.

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A C K N O W L E D G E M E N T

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P R E F A C E

The thesis entitled "Studies on the role of *Drosophila busckii* in the development of fruit rot of tomato caused by *Aspergillus niger*" is in two parts. Part I deals with the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of an insect, *Drosophila busckii* thus elucidating some information on the role of the insect in the development of fruit rot of tomato; and the effect of extracts of nineteen plant species on fruit rot of tomato (*Aspergillus niger*) in the presence of the insect (*Drosophila busckii*); the Part II deals with the effect of extracts of nineteen plant species when incorporated in the feed on egg laying, hatching of larvae, pupation and development of adult flies. Since the flies lay eggs on over ripe fruits, the effect of extracts of plants which were tested in part I was determined on development of flies and the results are given in part II.

PART I

CONTENTS

CHAPTER I

INTRODUCTION

Weather, insects and plant diseases are the three great natural hazards of crop production. The interaction of the two of the three and sometimes of all the three is so great and complicated that it becomes difficult to determine the actual origin of the trouble. Therefore, the importance of any of the three cannot in any way be undermined.

Plant diseases have been reported to take heavy toll of crops every year. Wood (1953) reported that stem rust of wheat alone caused losses in wheat to about $ 400,000 annually in one country alone. The bacterial wilt of tobacco in one county of North Carolina of the USA resulted in the loss of tobacco worth of 1-2 million dollors annually in the early part of the present century. By and large, in the USA the average annual loss from plant diseases is estimated to be about 3 billion dollors. It is difficult to enumerate all the figures of losses due to diseases of standing crop, but even after the crops have been harvested the products are attacked by destructive microorganisms. Succulent fruits and vegetables are more prone to rotting during transportation and storage. This is all the more true in developing countries where proper storage facilities are lacking. Although exact losses due to storage diseases of succulent crops are not

available but figures on other crops would indicate that they are of no less importance (Stakman and Harrar,1957). An average of about 2 percent of wheat harvested in the USA is deteriorated in storage. In 1951 the loss of winter wheat in storage was estimated to be between 5-10 percent of the crop (Stakman and Harrar,1957).

Exact estimates of losses of plants/products due to insects are not available but it is certain that they take heavy toll of the standing crop as well as plant products. The insect by providing injury to shoot reduces the photosynthetic area of the crop and impair the quality of the fruit (Metcalf and Flint,1973). The transmission of viruses by insects is well established (Takami, 1901). Thomas and Ark, (1934) observed that flies and ants initiated primary infection by carrying the fire blight bacteria from hold over, cankers to blossoms. Keitt and Ivanoff,(1941) reported that bees affected 49 percent transmission of fire blight bacteria during their visit to flower in search of nectar. Transmission of this organism has also been observed by tarnished plant bugs,wasps, aphids, bark beetle, codling moth and yellow jackets. Ark and Thomas,(1936) showed that the pathogen could survive in the intestinal tract of the three species of flies, Drosophila melanogaster, Musca domestica and Lucilia sericata.

List and Kreutzer,(1942) provided evidence that ring rot of potato caused by Corynebacterium sepedonicum could be transmitted by grasshopper (Melanoplus differentialis), black blister beetle (Epicauta pennsylvanica) and the potato beetle (Leptinotarsa decemlineata) which fed on diseased plants and tumors.

In nature fungus and insect develop some kind of relationship which has been categorised as follows :(Austwick, 1958).

1. Infection incident to pollination.
2. Infection through traumatic injury.
3. Internal and external contamination of the insect as a result of feeding on fungal masses.
4. Fungi developing on insect exudates.
5. Infection through feeding and oviposition wounds.
6. Infection through feeding punctures.
7. Infection resulting from symbiotic association between insect and fungus.

In all these categories of relationships the insect either acts as a carrier or provides a wound for the entrance of pathogen. The literature has been reviewed by various workers.(Leach , 1940& Carter , 1962). The insects are well equipped to act as vectors, because most of them depend on plants for food. They are generally active, and their body

bristles enable them to carry many pathogen spores externally (Austwick , 1958). In addition, many plant pathogens can survive and even multiply inside the insects (Mercier, 1911). Insects during pollination feed without wounding the plant and act merely as vectors of fungi. Botrytis anthophila sporulates only on anthers of red clover and is spread by bees while Ustilago violacea is spread by nocturnal moth. A small hymenopteran, Blastophaga psenes L., ensures an unusual method of pollination called caprification and causes an internal rot of fig by carrying spores of Fusarium moniliforme Var. fici from infected to healthy fruits (Caldis , 1927).

Insects have also been found to carry fungi on their mouth parts and introduce them into plants during feeding. Transmission of Nematospora gossypii causing stigmatomycosis of cotton probably depends on cotton stainers (Dysdercus spp.) (Frazer , 1944). Plant bugs are noted for the injury they cause when feeding on plants. Leach (1940) suggested that the necrosis may not always be caused by toxic saliva of the insect but also by associated fungi of Nematospora type.

Larvae of the corn borer Pyrausta nubilalis have been reported to disseminate several pathogenic fungi within maize plants (Christensen and Schneider, 1950). Wasp is reported to inoculate, Stereum sanguinolentum (Fr.) in conifers causing heart rot (Cartwright , 1938). Parkin (1942) pointed out that larvae of the insects feed on hyphae and carry the fungus in

their hypopleural organs. Crickets of genus, Oecanthus have been shown to carry Leptosphaeria coniothyrium causing tree cricket canker of apple and Cane blight of raspberry (Gloyer and Fulton, 1916). Azalea flower spot (Ovulinia ozaleae, Weiss) is spread by atleast eleven species of bees, three species of ants, and a thrip (Heterothrips azaleae), (Smith and Weiss, 1942). The most common insects visiting this fungus bearing stickymass are Diptera and few Coleoptera. Mercier (1911) observed the spores of fungus externally on a fungus gnat (Sciara thomae L.) which passed through the intestinal tract in a viable condition. Yarwood (1943) found that while feeding on the mildew colonies on rose and grape leaves, the thrips could transmit the conidia. Yamamoto (1951) reported that fragments of the fungi pass through the intestinal tracts of insects, i.e,, flies, wasp, bees and lady bird beetles. Scott and Ayres (1910) reported that 93% of fruit infection could be traced to curculio wounds, although brown rot was often present in the absence of curculios. Ogawa (1957) reported that C. hemipterus L., were contaminated with spores of Sclerotinia fructicola during feeding on diseased patches and was able to transmit the organisms to naturally wounded peaches.

The association of the weevil, Rhyncites baccus (L.) with conidia of the Monilia spp. has been studied extensively by Stojanovic (1956). Conidia of the fungus were found on

legs and lower surface of abdomen. Gruenhagen et al. (1947) presented adequate evidence of transmission of fungal spores by insects and suggested that in addition to acting as vectors, the insects also served as agents to weaken the twigs and render them more susceptible to fungus entry. Griswold (1953) observed that Drosophila species could move spores from fungus mats to wounds. Later Griswold (1956) showed that D.melanogaster transmitted the fungus after feeding on fungus hyphae. The most efficient relationship is where the insect transmits the pathogen from plant to plant and also makes the wound through which infection takes place. This kind of relationship has been observed between dutch elm disease and elm bark beetles (Leach , 1940).

Tomatoes are used for vegetables and for salad and are consumed throughout the world. From harvest to market the fruits are stored under various conditions and are exposed to hazards of various insects and pathogens. Drosophila busckii, an insect of the order, Diptera is commonly found visiting the vegetable shops and warehouses in Aligarh. Ripened fruits in the shops have also been found rotting as a result of infection with Aspergillus niger. It was not known whether there is any relationship of the insect which is a frequent visitor of vegetable shops in the development of fruit rot by A. niger. Therefore, in the present studies an attempt has been made to work out systematically the role of insect in the development of fruit rot. The following aspects have been studied:-

1. Survey of insects found in vegetable shops.
2. Survey of fungi found associated with tomato fruits sold in Aligarh market.
3. Pathogenicity of Aspergillus niger on tomato fruits.
4. To detect the presence of inoculum on the body of insect.
5. Effect of bagging of fruits in plants on fungi on the fruits.
6. Effect of temperature on the development of fruit rot of tomato caused by A. niger in the presence of D. busckii.
7. Effect of relative humidity on development of fruit rot caused by A. niger in the presence of D. busckii.
8. Changes in ascorbic acid (vitamin C) content in tomato fruits inoculated with A. niger and fed by D. busckii.
9. Changes in amino acid content in tomato fruits inoculated with A. niger and fed by D. busckii.
10. Studies on control of fruit-rot caused by A. niger in the presence of D. busckii.
    - 10.1 Effect of treatment with ethanol.
    - 10.2 Ethanolic leaf extract of Lantana camara, Mentha arvensis and Ocimum sanctum.
    - 10.3 Dry powder of leaves and extract of leaves of neem in water and alcohol.
    - 10.4 Effect of water extracts of certain plant on the development of fruit rot caused by A. niger in the presence of D. busckii.

10.5 Effect of latex of Euphorbia hirta and Calotropis procera on development of fruit rot caused by A. niger in the presence of insect D. busckii.

10.6 Effect of growing tomato seedlings in soil treated with different oil cakes and bavistine on the development of fruit rot when inoculated with A. niger in the presence of D. busckii.

CHAPTER II

REVIEW OF LITERATURE

According to Pennington (1913) the New York Board of Health rejected 12,000,000 lbs of different fruits because partly of poor quality and partly attacked by microorganisms. Since then there has been growing awareness amongst the scientists and growers of the losses of fruits and vegetables due to post-harvest diseases. Adams (1916) reported that 25 per cent of the fruits and vegetables were deteriorated due to microorganisms during storage.

Stakman and Harrar (1957) listed *Aspergillus* , *Penicillium*, *Septoria*, *Alternaria*, *Helminthosporium*, *Macrosporium*, *Cladosporium*, *Hormodendron*, *Botrytis* and *Rhizopus* as the common genera of fungi causing blemishes, moulds and rots of various succulent fruits. On soft fruits such as berry fruits, currants, drupes, the most important post harvest pathogen is greymold fungus, *Botrytis cinerea* (Edney, 1964; Dennis and Mountford, 1975; Davis and Dennis, 1977; Mason and Dennis, 1978), followed by *Mucor*, *Rhizopus* (Lowings, 1956; Dennis, 1978; Mason and Dennis, 1978) and *Cladosporium* spp. (Cappellini *et al*., 1961; Dennis, 1975; Mason and Dennis, 1978).

Jarvis and Borecka (1968) pointed out that the fruits contract to infection of Botrytis cinerea at the "open flower" stage and that infections remain latent. This shows that, it is not essential that fruits should become infected during transit and storage.

In England Preece (1967) reported that apples are infected by Gleosporium spp. and Monilia fructigena during storage, while Swinburne (1970) observed that Nectria gallignea and Penicillium expansum are main pathogens during storage. In France, Bondoux (1967) had suggested that Gleosporium spp., M. fructigena, Botrytis cinerea and Penicillium spp. are main organisms involved in the losses of apples during storage. In Newzealand, Cooper and Padfield (1965) also listed 15 different species of fungi causing rotting of apples. Surveys carried out in Poland (Ostrowski, 1971), East Germany (Katschinski, 1974) and Yugoslavia (Babovic et al., 1979) indicated Penicillium, Monilia and Gleosporium amongst important causes of rotting of apples.

Botrytis cinerea is also a major cause of post-harvest fruit rotting of tomatoes (Chastagner and Ogawa, 1979; Dennis and Davis, 1980). Other fruit rotting fungi on tomato include Alternaria spp. (Bartz, 1972; Pearson and Hall, 1975; Dennis et al., 1979), Stemphylium spp. Fusarium spp., Cladosporium spp. and Rhizopus stolonifer (Dennis, 1981). There are many more reports of fungi deteriorating fruits

during storage and the literature has been reviewed by Dennis (1983).

In India, also large number of fungi have been reported from fruits including succulent ones (Vyas et al., 1976; Sohi, 1983; and Bhargava and Arya, 1983). Agarwala and Sharma (1968) isolated as many as 15 different fungi from apples. Trichothecium roseum was reported from apple during processing (Sreekantiah et al., 1974). P. expansum , G. cingulatus, T. roseum and R. stolonifer were commonly found attacking apples during the survey of markets, godowns and canning centres (Kaul and Munjal, 1980) and Aspergillus candidus during storage (Thind et al., 1976). Koltz and Fawcett (1948) added Fusarium spp. in the list of fruits attacking citrus. Aschochyta caricae has been reported to cause serious losses of papaya (Chaudhury, 1950). Cucurbitacious fruits were damaged by Alternaria alternata in storage (Laxminarayana and Reddy, 1976). Kale and Raut (1985) observed that mandarin oranges(Citrus reticulata) were badly damaged by Colletotrichum gloeosporioides and Fusarium fusiarioides. Prakash et al.,(1974) pointed out that anthracnose of Cucumis melo var. momordica caused by Colletotrichum capsici damaged the fruits even in storage. Snake gourd (Trichosanthes anguina) has been found to be infected with Alternaria tenuissima (Singh, 1974). Gangopadhyay and Sharma (1976) observed pumpkin (Cucurbita moschata) infected with Rhizoctonia solani and Fusarium oxysporum causing spongy rots. Sahu Kritagayan and Singh (1980)

reported that fruit rots of parwal and pointed gourd (Trichosanthus dioica) were caused by a number of fruit rotting fungi. Momordica dioica fruits were found infected by Fusarium semitectum in the market. Thirumalachar (1953) Kapoor and Chauhan (1974) and Singh (1975) observed that Macrophomina phaseoli caused charcal-rot and Ashy stem blight of wide variety of fruits such as those of legumes, water melon, potato and papaya. Grewal (1954) and Tandon (1967) pointed out that Ascochyta caricae, Gloeosporium caricae and Colletotrichum caricae attacked papaya fruits. Aspergillus awamori, A. fumigatus, A. flavus, A. niger and Dreschlera rostrata (Srivastava and Tandon, 1971) and Pestalotiopsis versicolor and P. glandicola (Vayas and Kanwar, 1974 and Agarwal and Hasija, 1974) have been reported from pomegranate and T. roseum from banana (Srivastava and Tandon, 1971). Pestalotiopsis versicolor was also found damaging fruits of Acharas sapota (Sohi, 1983). Dhingra et al.,(1980) also found P.glandicola infecting sapota. Aspergillus awamori, A. fumigatus, A. flavus, A. niger and Dreschlera rostrata. (Philip ,1979), A. nivens and A. versicolor (Sharma et al., 1981) were reported to cause fruit rot of pomegranate. Jamaluddin, et al.,(1972) observed Gliocephalotrichum bulbilium commonly damaging fruits and vegetables in storage. The fruits of Litchi chinensis have been found badly damaged by Aspergillus flavus, A. niger, A. nidulens, A. quadrilineatus,

A. variecolor, Botryodiplodia theobromae, Colletotrichum gloeosporioides, Cylindrocarpon tonkinense and Pestalotiopsis sp . (Prasad and Bilgrami, 1973). The tubers of Dioscorea alata were badly damaged by Penicillium sclerotigenum. (Agarwal and Gupta, 1973). Cylindrocladium scoparium was found infecting apple (Malus sylvestris), guava (Pisidium guajava), Aonla (Pyllanthus emblica) pea pod (Pisum sativum). Fruits of Aonla (Pyllanthus emblica) were also found infected with Phoma putaminus. A pre and post-harvest fruit rot disease of Aonla was reported to be due to Calophyllum inophyllum (Wadia et al., 1984). Subrahmanayam and Sarma (1974) reported that the severe fruit rot of Emblic myrabolam was caused by Penicillium funiculosum, Colletotrichum dematium and Fusarium semitectum. Saharan and Gupta (1974) observed C. dematium, F. semitectum and F. moniliforme infected soybean pods both in field and storage. Ripe fruits of strawberry were damaged by Colletotrichum fragariae (Singh , 1974). Sclerotium rolfsii has been reported to be a serious pathogen of the standing vegetable crops but if fruits infected are stored, the damage increases. In a study on the host-range of the fungus, the chillies were found highly susceptible, banana and lemon moderately and guava resistant. Kaul and Lall (1975) while making a survey of the post-harvest diseases of fruits and vegetable found that Penicillium italicum was the most frequently found on different fruits. The affected fruits were found to be mummified under low humidity followed

by P. digitatum, Aspergillus niger, Geotrichum candidum and Fusarium sp.). Chaurasia (1980), while making extensive studies reported that a rot of Agele marmelos (bael) was caused by Phoma glomerata. Dhingra et al., (1980) pointed out that Jambolan or Java-Plum (Syzygium cuminii) has been reported to be damaged by Pestalotiopsis palmarum, Penicillium expansum, Rhizopus stolonifer, Fusarium semitectum, A. niger, Curvularia lunata, Colletotrichum gloeosporioides and Sclerotium rolfsii during post-harvest stages (Wadia and Manoharcharya, 1982) and plums (Prunus domesticana) with Geotrichum candidum (Saikia and Puzari, 1982). Saxena et al., (1983) pointed out that bean pods (Dolichos lablab) were commonly found infected with Geotrichum candidum exhibiting white powdery appearance of spores and conidiophores of the pathogen. Fruits of guava (Pisidium guajava) were found rotted due to Fusarium solani (Chakarbarti, 1983). In addition to the above Aspergillus niger, Rhizoctonia solani, Colletotrichum gloeosporioides, Botryodiplodia theobromae, Erwinia sp., Rhizopus stolonifer, R. oryzae and Choanephora cucurbitarum also caused softrots and A. fumigatus, Penicillium sp., P. multicolor, Cladosporium sp., Fusarium equiseti and F. oxysporum as dry rot of guava (Adisa, 1985). Singh and Kainsa (1983) concluded that Aspergillus niger, A. rugulosus, A. teereus, Penicillium, Chrysogenum and Saccharomyces sp. are among those causing highest decay of fruits in the market (Sohi, 1983).

Sugunakar Reddy. and Chandra Reddy (1983) reported that fruit rot of grapes (Vitis vinifera) was caused by Greeneria uvicola and by Myxosporium pisidii. Mandal and Das Gupta (1984) observed that fruits of ber (Zizyphus jujuba) were found infected with Myxosporium pisidii. Tomato fruits, like other vegetables, are also subjected to hazards of post-harvest. Jamaluddin et al., (1974) pointed out that Cylindrocladium rot was found to be more destructive post-harvest disease of tomato. They were also found highly susceptible to S. rolfsii (Singh et al., 1975). During the survey of Aligarh markets the fruits were found rotted with different species of Aspergillus, Penicillium and Rhizopus in the shops and warehouses.

Post-harvest damages are influenced by various environmental factors, particularly those of storage. Harter and Weimer (1922) isolated 11 species of Rhizopus from fruits and vegetables with each species requiring different temperatures for infection and spread. R. chinensis required 35°C while R. oryzae, R. maydis, R. tritici, R. delemar, R. nodusus and R. arrhizus 30°C and R. artocarpi and R. nigricans 20-25°C for maximum damage. Adams (1923) reported that the development of Penicillium glaucum, Penicillium sp., Venturia inaequalis, Gleosporium, Cladosporium pleospora, and Botrytis cinerea was best at 25-32°C during storage with no growth at 34°C. Smoot and Segall (1963) observed that

there was no development of anthracnose of mango caused by Colletotrichum gloeosporioides at 130-135°F. Strawberries are even infected by B. cinerea and M. piriformis at a temperature of 0°C (Dennis and Cohen, 1976). However, fruits do not become infected by R. stolonifer at this temperature. Tandon and Ghosh (1962) pointed out that optimum temperature for the development of Alternaria tenuis on pears ranged between 20-25°C. Botryodiplodia theobromae and Pestalotia sapotae caused highest damage of Sapodilla at 25-30°C during storage. Further Rhizopus rot of papaya was initiated at 15°C but the development of rot was highest at 25-30°C. (Tandon and Misra, 1969). Highest rotting of litchi fruits by Aspergillus niger, A. flavus, A. quadrilineatus, A. nidulens, A. variecolor, B. theobromae, Colletotrichum gloeosporioides, Cylindrocarpon tonkinense and Pestalotia sp. occurred at 15-25°C (Prasad and Bilgrami, 1973) while that of tomato by Alternaria solani and A. tenuis at 25°C (Mehta et al., 1975a). Pathogens on mango failed to develop at 10°C (Banerjee and Rao, 1933; Tindale and Trout, 1936; Wardlaw and Leonard, 1936; Cheema et al., 1939; Karmarkar and Joshi, 1940; Sethi, 1943 and Mukerjee, 1961). Tandon (1967) reported that decay of banana fruit caused by Gleosporium musarum was least when stored at 10°C, while the decay of mango and banana due to B. theobromae was low at 5-10°C and a temperature above 35°C.

Relative humidity of the air is equally responsible for causing rotting of fruits and vegetables. The development of various fruit rots caused by Aspergillus niger, A. flavus, A. nidulens, Alternaria tenuis, A. solani, Botryodiplodia theobromae, Cladosporium herbarum, Curvularia lunata, Geotrichum candidum, Rhizopus nigricans and R. arrhizus was highest at a relative humidity ranging from 80 to 100 per cent (Dastur, 1921; Park, 1930; Chaudhury, 1950,1955; Sattar and Hafiz, 1953; Tandon and Singh, 1969; Prasad and Bilgrami, 1973; Mehta et al., 1975).

Moderate temperatures and high humidity have been found to influence the pear rot caused by Sclerotium rolfsii (Sumbali and Mehrotra, 1983). Singh et al., (1983) pointed out that tomato fruit rot by Cladosporium oxysporum was highest at 25-30°C and 95 per cent relative humidity.

During the course of infection and development of post-harvest fruit rots changes have been observed in carbohydrate, amino acid, ascorbic acid, organic acid contents (Ulrich, 1958; Biale, 1960; Wood, 1960; Tandon,1967; Srivastava, 1969; Tandon, 1970; Coursey, 1972; Bhargava and Arya, 1983). A decrease in sugar content was observed in pine apple, banana, mango, sapotas and musambi infected with Botryodiplodia theobromae (Bhargava, 1962); in banana with Fusarium oxysporum f. cubense (Chandra and Tandon, 1963); in apple with Hendersonula toruloidae and Aspergillus niger (Mc Combs and Winstead, 1964); in cucumber with

Pythium aphanidermatum; in guava, papaya, sapodila and banana with Pestalotia psidii, Phoma psidii, Gleosporium sp. Colletotrichum papayae, Fusarium sp., Pestalotia sapodilla and B. theobromae (Ghosh et al., 1964); and in blue berry fruits with Glomerella cingulata (Stretch and Capellinii,1965).

There was a decrease in the content of glucose, fructose and sucrose in guava infected with Macrophomina allahabadensis (Kapoor and Tandon, 1967) in pomegranate, guava and musambi with Aspergillus niger., in banana and papaya with Rhizopus stolonifer; in tomato with Phoma sp. (Aulakh et al., 1970b); in Chilli with Choanephora cucurbitarum (Chahal and Grover, 1972); in banana with Chochliobolus spicifer and Alternaria alternata (Prasad, 1974); in tomato with Cylindrocarpon scoparium, Myrothecium roridum and Colletotrichum sp., and in Chilli with Rhizopus stolonifer (Tandon et al., 1974), papaya with Phomopsis caricae papayae (Dhingra and Khare, 1975); Yam with Sclerotium rolfsii (Ogundawa et al., 1975); tomato with Alternaria solani and A. tenuis (Mehta et al., 1975); musambi fruits with Botryodiplodia theobromae (Ali, 1976) and apple with Clathridum corticola (Thind et al., 1977).

Changes in amino acid contents with reduction in total amino acid or production of new amino acids have been observed in cucumbers infected with Pythium aphanidermatum (McCombs and Winstead, 1964), melons with Colletotrichum langenarium(Touz'e, 1964), blue berry with Glomerella cingulata

(Stretch and Capellinii, 1965); papaya with Gleosporium papayae, Colletotrichum papayae, B. theobromae and R. nigricans; banana with B. theobromae and Phoma psidii, Pestalotia psidii and Gleosporium psidii (Tandon, 1975); mango, guava and pomegranate with Aspergillus niger; tomato with Dreschlera australiense (Kapoor and Tandon, 1960); mango, sapota and citrus with B. theobromae (Tandon, 1967; Srivastava and Tandon, 1969a); tomato with Phoma sp. (Aulakh et al., 1970a), Chilli with Choanephora cucurbitarum (Chahal and Grover, 1972); tomato with Cylindrocarpon scoparium, Myrothecium roridum and Colletotrichum sp., chilli with R. stolonifer (Tandon et al., 1974), tomato with Alternaria tenuis and A. solani (Mehta et al.,1975b); apple with Clathridium corticola (Thind et al., 1977) and cucurbits with Pythium butleri (Singh and Chohan, 1977).

Ascorbic acid (vitamin C) is an important constituent of fruits and vegetables in general and tomato in particular. Infection of fruits with fungus in general resulted in the reduction in total ascorbic acid content, (Bhutani, 1946; Kamat et al., 1952), such as in guava fruits infected with Pestalotia psidii, Phoma psidii and Gleosporium psidii (Ghosh et al., 1965) and Aspergillus niger (Singh and Tandon, 1971) papaya with R. nigricans, B. theobromae, Colletotrichum papayae and Gleosporium papayae (Ghosh et al., 1966) with Alternaria tenuis, Chaetomium globosum, Curvularia lunata,

Cylindrocarpon tonkinense, Fusarium oxysporum and Helminthosporium speciferum (Prasad and Verma, 1976; Prasad, 1977; Prasad and Prasad, 1977); apple by several fungi (Bisen, 1974; Agarwal and Bisen, 1976; Agarwal and Nema, 1979); and in musambi and orange (Agrawal and Ghosh, 1979) in Zizyphus jujuba (Prasad, 1980); tomato with Dreschlera austrialianse (Kapoor and Tandon, 1969); Chilli with Choanephora cucurbitarum (Chahal and Grover, 1972) and tomato with Cylindrocarpon scoparium, Myrothecium roridum and Colletotrichum sp., Chilli with R. stolonifer (Tandon et al., 1974; Jamaluddin et al., 1975). Reduction in ascorbic acid content due to infection differs from the type of fruit and also in amongst variety to variety in a fruit. In guava var. Apple-colored infected with B. theobromae, there was a reduction of 10.8 per cent of ascorbic acid, whereas there was practically no reduction in safeda variety (Srivastava and Tandon, 1966b). Tandon (1970) observed a reduction of 81.3% in ascorbic acid content in papaya fruits infected with A. niger and 92.0% in musambi infected with B. theobromae. Singh and Tandon (1971) observed that in healthy and Aspergillus spp. infected guava fruits there was loss in the vitamin C, with increase in duration between 6-12 days after infection. Infection of litchi, (Litchi chinensis sonn) China and Shahi a remarkable decline in ascorbic acid content was observed during 6-10 days of pathogenesis of Aspergillus flavus, Aspergillus niger, A. variecolor, A. nidulens and A. quadrilineatus (Prasad and Bilgrami, 1979). The most affective in this regard was

A. flavus followed by the forms in the order mentioned above. Jamaluddin et al., (1975) reported that 86.7% loss in ascorbic acid content of the tomato fruits of the same variety after ten days of infection of Myrothecium roridum. Reddy et al., (1980) observed 91-100% loss in its amount in tomato fruits infected separately with Phoma exigua, Rhizoctonia solani, Stemphylium vesicarium and Nigrospora oryzae. Gangawana and Datar (1978) found decrease in ascorbic acid in the leaves of tomato CVS. susceptible to Alternaria solani. The extent of decrease was greater in highly susceptible cultivars.

Bisen (1974) also found complete loss of vitamin C after twelve days of infection of Aspergillus niger in apple varieties 'Kesari' and 'Edward'. Agarwal and Nema (1979) found almost similar level of diminution in ascorbic acid content in the fruits of apple Cultivars American and Delecious during three to twelve days of infection of Alternaria alternata. The loss, however, was not as drastic as found in apple fruits infected with Aspergillus niger within 10 days of separate infection of Pestalotia anonicola, Stachybotrys sp. and Trichoderma viride (Chawdhury et al., 1980). In Citrus medica (Lemon) fruits infected with Colletotrichum gloeosporioides the content of ascorbic acid diminished almost to half and Citrus sinensis (Musambi) more than half after twenty days of infection (Agarwal and Ghosh, 1979). Reddy et al., (1984) found gradual loss in ascorbic acid content both

in healthy and Aspergillus niger infected fruits of lemon Citrus aurantifolia. However, the diminution was more rapid in infected fruits.

Sinha and Singh (1984) reported reduction in ascorbic acid in pear fruits to be up to 80.6 and 87.3% after 7 days of infection of A. flavus and A. parasiticus respectively. Similarly,in fruit pulp of peach infected with Rhizopus stolonifer ascorbic acid content diminished to the extent of two and half times to that of healthy one (Singh and Prashar, 1984).

Attempts have been made to reduce the losses due to post-harvest diseases. The literature pertaining to chemical treatments, for the control of post-harvest diseases has been reviwed by Eckert and Sommer (1967). Boric acid immersion has been used for citrus fruit (Perrel and Laspes, 1949) and mango against anthracnose (Clara, 1928); formalin as wash for mango (Banerjee and Karmakar, 1934) and decay of tomatoes (Ramsey and Bailey, 1934); Sodium bicarbonate for controlling post-harvest fungi on citrus (Childs and Seighlar, 1946) and mercuric chloride on tomatoes (Stakman and Harrar, 1957). Meredith (1960a,b) obtained good control of post-harvest diseases of citrus by Alphanaphthalein acetic acid. DiMacro and Davis (1957) and Van Blaricon,(1959) could control certain post-harvest diseases of peaches by applying antibiotics such as Nystatin, Aureofungin and Pimaricin. Strawberry rots

caused by Botrytis sp. (Salunkhe et al., 1962), Gleosporium rot of banana (Meredith, 1960a,b) and Botrytis cinerea on grapes (Stessel, 1958) were controlled by applying antibiotics. Srivastava and Tandon (1969) obtained good control of Botryodiplodia theobromae on guava and Ananthanaryana and Seshdhari (1965) peach brown rot and anthracnose of banana by applying antibiotics. Becker et al., (1958) recommended cyclohexamide to control post-harvest rot of strawberries caused by Botrytis sp. and Rhizopus sp. Aureofungin has been effectively used to control Alternaria rot of tomato, Diploidia rot of mango (Dharmvir et al., 1966); Pythium rot of cucurbits (Sharma and Wahab, 1970,1971); Glomerella sp. and Aspergillus niger on various fruits (Laxminarayana and Reddy, 1974). Tetracycline has also been found very effective for controlling Colletotrichum gloeosporioides on guava (Sohi, 1983). There should be coordination of pre and post-harvest treatments to reduce ultimate losses during storage and marketing. Vineyards have been sprayed at bloom time with benomyl [methyl 1- (butylcarbamoyl)-2-benzimidazole carbamate] in combination with Captan (N-trichloromethyl-mercapto-4-cyclohexene-1,2-dicarboximide) or 2,6-dichloro-4-nitroaniline (DCNA) and then were dusted with captan or DCNA in the late summer and fumigated with (SO_2) after packing (Harvey and Uota, 1977). Application of benomyl as dormant, bloom and pre-harvest sprays reduced the level of decay before and after harvest. Decay was further reduced by post-harvest

treatment with wax emulsion containing DCNA or a combination of DCNA and benomyl (Wells and Gerdts, 1971a and 1971b). Post-harvest losses have also been minimised by host water treatment which is advantageous as it leaves no residue (Harvey, 1978). Papayas in Hawaii have been treated with hot water for controlling both fruit flies and decay caused by Colletotrichum sp. and Gloeosporium sp. Akamine and Arisumi (1953) treated fruits with hot water at 49°C for 20 min. for controlling anthracnose and other diseases. Akamine and Goo (1969) obtained good control of insects with fumigation. Akamine (1977) was also able to control storage decay of mangoes by treating them with hot water at 47°C for 20 minutes. Post-harvest losses of peaches have been reduced by hot water dips for 3.5 min at 49°C or 1.5 min at 54°C. Smith and Redit (1968) observed that a 2-3 min. exposure of fruits at 52°C effectively controlled fruit decay. Wells (1971) could reduce the decay of nectarines by 65-75% by dipping at 52°C for 1.5 min. He further observed that adding DCNA, captan or benomyl to hot water increased the effectiveness of the treatment. The exposure time to hot water was also reduced to 0.5 min. Wells (1972) reported that combination treatment allowed the concentration of fungicide to be reduced without loss of efficacy. Heated wax emulsions containing benomyl and DCNA provided more effective decay control.

Insects not only cause direct injuries to plants but also involved in tranmitting the pathogens. In transmitting the insects develop some kind of relationship with the host plant and the pathogen (Carter, 1962). Although considerable work has been carried out on direct injuries on losses to plants caused by insects (Leach, 1940 and Carter, 1962), but it is difficult to review them here. Therefore, losses done on vegetables are summarised in table, 1. Mills and Sinha (1971) observed that when specimens of Hypogastrura tullbergi were exposed to 43 species of soil fungi maintained at P.D.A. at different R.H., feeding and reproduction of the insect were observed on Alternaria alternata, Cladosporium sp. Cladosporioides sp., Bipolaristetamera sp. and Sporotrichum carmis. These fungi formed a mat around the insect body resulting in low reproduction. Feeding and reproduction of the insect were highest at 15°C. Fresa (1971) and Berisford and Taso,(1974) observed that the fungus Entomophthora grylli had a unique relationship which filled the body cavities of grasshoppers. The insects were not killed even after 12 or more days of infection but infected females were unable to oviposit. Williams and Dove (1972) pointed out that tubers were damaged by the larvae of Agrotis ipsilon and other soil inhabiting species such as mole crickets. Highest damaging was the larvae of Phthorimaea operculella. Bemisia tabaci has been observed on large number of vegetables and often involved in transmission of viruses (Carter, 1962). Herakly and Ezz (1970) observed highest infestation of white fly on

Table 1. Important insect pests reported from vegetables

Vegetable		Insect pests			Zone/area reported	References
Latin name	Common name	Latin name	Common name	Losses/Damages	Reported from	
1. Allium cepa	Onion	Thrips tabaci	thrips	Seed, crop and flower stalks affected (20-25%).	Punjab (India)	Thind and Jhooty (1982)
2. Artocarous heterophyllus	Jackfruit	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al.,(1975)
3. Brassica campestris	turnip	Thrips hawaiiensis	thrips	Flower showing dull and sickly appearance with brownish lesions on stamens and stylets. Heavily infested bloom were unable to form fruits.	H.P. (India)	Srivastava and Bhullar (1980)
4. Brassica campestris Var. capitata	Cabbage	-	Pale legume bug	Damage of inflorescence.	U.S.A.	Getzin, (1983)
5. Brassica oleracea	Celery	Euleia (Philophylla) heraclei	-	Internal changes (Parenchyma deteriorated) small leaflets.	Grand mount (France)	Leroi, (1974)
6. Brassica napus	Oil seed rape	Ceutorhynchus assimilis	Weevil	Seed yield affected.	Harpenden (U.K.)	Free et al.,(1983)
7. Brassica oleracea var. botrytis	Cauliflower	-	Cabbage moth	Damage to cabbage leaves.	Kostinbrod (Bulgaria)	Straka,(1983)
8. Brassica oleracea var. botrytis	Cauliflower	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al.,(1975)
9. Capsicum annum	Chilli	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al.,(1975)
10. Capsicum frutescens	Chilli	Scirthothrips dorsalis, Polyphagotarsonemus latus	thrips	Leaf curling and malformation.	Maharashtra (India)	Amin, (1979)

Contd...

Table 1 (Contd.)

Vegetable		Insect pests			Zone/area reported	References
Latin name	Common name	Latin name	Common name	Losses/Damage	Reported from	
11. Carissa carandas	Karaunda	(Unidentified) insect	-	Mature fruits rotted completely, remained attached to the trees as firm mummy or fall.	Allahabad (India)	Tandon and Kumar (1974)
12. Colocasia spp.	taro	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique, and French (Guiana)	Panis et al.,(1975)
13. Cucumis spp.		Diabrotica undecimpunctata	Beetle	Injured cucurbit leaves.	Lexington (U.S.A.)	Bergstrom et al., (1982)
		Acalymna vittatum	Beetle			
14. Cucurbita spp.		Diabrotica undecimpunctata	Beetle	Injured cucurbit leaves.	Lexington (U.S.A.)	Bergstrom et al., (1982)
		Acalymna vittatum	Beetle			
15. Daucus carota	Carrot	-	Fungus gnat larvae	Cavity spots in carrot.	Ithaca (U.S.A.)	Hafidh and kelly (1982)
16. Glycine max	Soyabean	Melanoplus differentialis and M. bivittatus	Grasshopper	Defoliation (100,50, 1-10 and 0%).	Arkanas (U.S.A.)	Muelleret al., (1980)
17. Ipomea batatas	Sweet potato	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al., (1975)
18. Lycopersicum esculentum	tomato	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al., (1975)
19. Momordica charantia	bittergourd	Thrips hawaiiensis	thrips	Flower showing dull and sickly appearance with brownish lesions on stamens and stylets. Heavily infested bloom were unable to form fruit.	H.P. (India)	Srivastava and Bhullar (1980)
20. Manihot esculenta	utilissima	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al., (1975)
21. Phaseolus vulgaris	bean	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al., (1975)
22. Solanum melongena	eggplant	Pseudococcus virgatus	mealy bug	Leaves are damaged, favour the entry of fungus.	Guadeloupe, Martinique and French (Guiana)	Panis et al., (1975)
23. Solanum melongena	eggplant	Henosepilachna vigintioctopunctata	beetle	Stunted growth and reduced yield.	Madurai (India)	Raj and Lakshmanan (1980)
24. Solanum tuberosum	potato	Phthorimaea operculella	Potato tuber moth, cut-worm (moth)	Damage of tubers and foliage. (40-45%)	Mauritius	Williams and Dove (1972)
		Agrotis ipsilon				
25. Vicia faba	Cowpea	-	Weevil	Seed weight affected.	Fortaleza (Brazil)	de oliveira et al.,(1984)

egg plant followed by potato, marrow, cabbage and tomato, broad beans (Vicia faba). Graham and Mcneill (1972) reported larvae of Sciarid Bradysia coprophila feeding on the roots of soyabean in green houses facilitating the entry of Pythium and Fusarium. Other fungi such as B. theobromae, Macrophomina sp. have also been reported to gain entry through wound caused by squirrel, bird such as wood pecker. Tandon and Kumar (1974) provided evidence that fungi such as Alternaria alternata causing rot of Carissa carandas gain entry through the wounds made by insects. Panis et al., (1974) concluded that scale insects not only provided wound but transmitted Penicillium spp. through feeding punctures.

Evans (1973) reported that phytophagous invertebrates and sugar feeding Diptera carried viable inoculum externally on their bodies and mouth parts. The inoculum was also found to be borne internally, because in some cases presence of spores was detected in faeces. Kmitowa (1973) pointed out that Paecilomyces farinosus was more pathogenic to larvae of Galleria mellonella, when it has been cultured in presence of Penicillium spp. He concluded that species of Penicillium produced a stronger unfavourable effect on insects as compared to Fusarium or Aspergillus. However, Beauvaria bassiana was found to be less pathogenic to larvae in the presence of certain soil fungi.

Pathogenicity of these fungi is influenced by various internal and external factors, such as culture condition, feeding habits of the target species and habitat conditions. Nair and Mcewe (1973) observed that laboratory cultures of Hylemya brassicae were found infected when they were exposed to diseased plants. Such flies survived for a maximum period of 5-7 days after the appearance of characteristic abdominal hole through which conidia of the fungus were discharged. Grubs and insects were found to contain spores of certain plant pathogenic species of fungi (Deshpande et al., 1974). Coroso et al., (1975) reported that Hemiptera were involved in the dissemination of the fungus Nematosphora coryli the causal organisms of yeast-spot of soyabean. Coyle (1975) was able to isolate fungi belonging to 4 families and 18 species from the integument, fore gut, midgut, hind gut and faeces of Reticulitermis flavis. Of these some of them were plant pathogens. Jotwani (1976) recorded several insects visiting ergot infected fields but transmission of conidia was affected by only two insect species., viz. plant bug (Dolycoris indicus) and black ant (Camponotus) sp. Dakwa (1977) concluded that, fungi which were disseminated by insects are structurally and ecologically well adapted to insect transmission. Moreover, these fungi have also been isolated from various parts of their body which is an indication of the insect acting as vectors. The transmission of fungi through insects was also observed by Yey and Fapgues (1977).

in Beauvaria bassiana transmission through Leptinotarsa decemlineata. The insect during feeding ingested spores which remained unaffected by intestinal fluid (Murakoshi et al., 1977). Hasan (1982) provided evidence that spores of Colletotrichum graminicola were disseminated by Locusta migritoria. He further found that the spores remained viable even after passage through the intestine. Bergstrom et al., (1982) observed a relationship of gummy stem blight caused by Didymella bryoniae (Mycosphaerella melonis) in cucurbits with the incidence of beetles. Further necrosis in cucurbits has been reported to be caused by the interaction of these organism i.e., powdery mildew fungus, insect and Didymella bryoniae. Any one of these is unable to cause necrosis. Prakash and Kauraw (1982) observed that combined infestation by Sitotroga cerealella, Rhyzopertha dominica, Sitophilus oryzae, Tribolium castaneum, Oryzaephilus surinamensis and Latheticus oryzae with mites (Tyrophagus sp., Acaropsisdocta [Acaropsellina docta] and Pyemotes ventricosus caused heavy reduction in quantity, quality and viability of the grains. Sharma et al., (1983) and Grillo and Alvarez (1983) observed that ergot pathogen is transmitted by Leptoglossus gonagra and Nezara viridula during their feeding on hosts. Yang et al., (1983) reported that adults of Cylindrocopturus adspersus (Sunflower stem weevil) and C. adspersus carried the spores of M. phaseolina externally and transmited them to sunflower during oviposition. Swincer (1984) confirmed

that insects while feeding on lettuce, peppers and lupin not only caused direct injury but also transmitted the spores of pathogens. (Vozzo , 1984) Larvae of Curculio sp. Melissopus latiferreanus, Ephestia sp. (Contaiminent) and Valentinia sp. were found to act as vectors of Fusarium solani and Epicoccum purpurascens on various Quercus spp. Verma and Pathak (1984) while investigating the pathogenicity of ergot of pearl millet reported the association of eight species of insects. All of them were found to be contaminated with conidia of Claviceps fusiformis. Amongst the various insects Apis indica (A. cerana indica) and Tabanus rubidus carried the heaviest conidial load. The presence of witches broom organism was detected in the salivary glands of the insect vectors specially leaf hoppers fed on the diseased plants. Kulhavy et al., (1984) observed an association of Dendroctonus ponderosae and Pityogenes fossifrons and Armilaria mellea (Armillariella mellae) with beetles, which might probably help in the development of rot diseases. Moser (1985) observed Tarsonemus spp. acting as vector for Ceratocystis sp. Haung and Harper (1985) pointed out that where alfa-alfa leaves infected with Verticillium albo atrum were fed to leaf chewing grass hoppers (Melanoplus sanguinipes) and (M. bivittatus) to determine survival of the pathogen through their digestive tracts and when grasshopper faeces contaminated with V. albo atrum were burried near roots of alfa-alfa seedlings, 13-20.8% of plants became infected and developed

wilt symptoms after 6 week, thus confirming their survival through intestinal tract of Alfa-alfa leaf cutter bee. Megachile rotundata was found to act as a dispersal agent for Verticillium albo atrum (Haung et al., 1986). In these studies conidia were found to be present in the cuticle depressions of the abdomen around the mouth and various other parts of the body. Sengonca and Leisse (1984) reported that Scolylids, Scolytus scolytus, S. multistriatus and S. pygmaeus, Pteleobius vittatus and P. kraatzi transmitted the pathogen, Ceratocystis ulmi causing Dutch elm disease. However, a considerable increase in the infestation of younger trees was observed during the study.

It is clear from the brief review that although considerable work has been carried out on insect-plant and insect-fungus-pathogen (insect acting as vector) relationship but practically nothing is known about the development of post-harvest rots in the presence of insects. Therefore, the present investigations were undertaken.

CHAPTER III

MATERIALS AND METHODS

3.1 A survey was undertaken for various insects found in shops; and fungi associated with rotting of tomato fruit, in Aligarh market during three seasons viz., rainy, winter and summer. During rainy season the survey was undertaken in July; winter in Dec/Jan; and summer in June which are normally considered peak of the seasons. The survey was spread over the whole period so selected.

Insects were collected by using wire-nets fitted in a ring of 20 cm iron rod with a handle. Insects so collected were stored in jars fitted with muslin cloth and were brought to laboratory. They were identified and their numbers were counted. The frequency of occurrence was calculated as follows:-

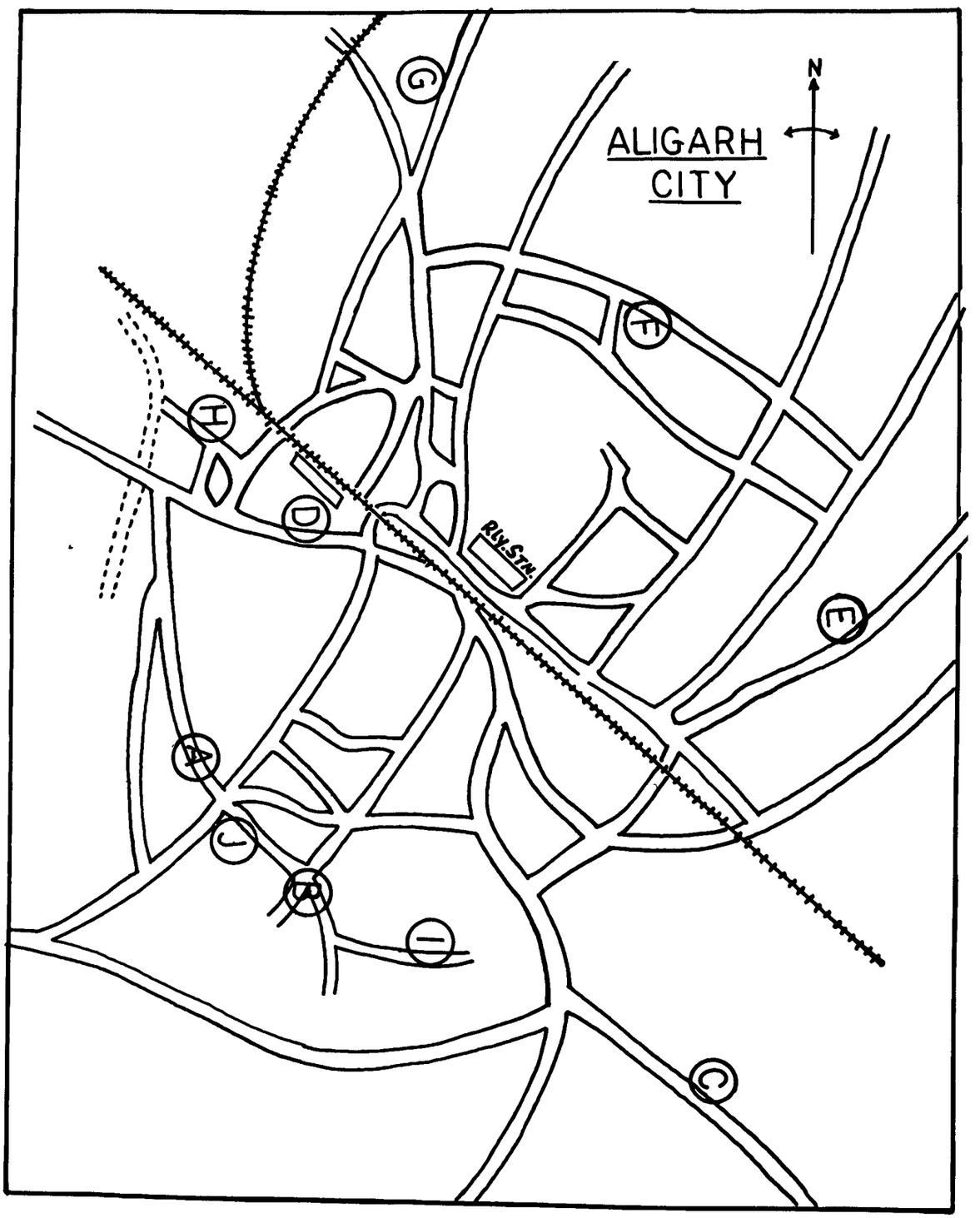
$$\text{Frequency} = \frac{\text{No. of insects of a species}}{\text{Total number of insects}} \times 100$$

In all five surveys were made, spread over the whole period and the mean was calculated.

Tomato fruits showing the sign of rotting were collected from shops and were brought to the laboratory in sterilized polythene bags. The rotted portions of the fruits were plated

Fig.1 Map of Aligarh city showing the sites of markets where collection was made.

A. Raghubir puri
B. Sabzimandi
C. Agra road
D. G.T.road
E. Vishnupuri
F. Dodhpur
G. Shamshad market
H. I.T.I. road
I. Jaiganj
J. Barhaduari


ALIGARH
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directly on the petriplates containing potato-dextrose-agar* containing rose bengal. Plates were incubated at 25°C. Fungi appearing around the tissue were isolated and identified. There were 10 plates for each locality. Frequency of occurrence of different fungi was calculated as follows:-

$$\text{Frequency} = \frac{\text{No. of plates containing a particular fungi}}{\text{Total number of plates}} \times 100$$

Since, the frequencies of Drosophila busckii amongst insects and Aspergillus niger amongst fungi were highest in all the localities in different seasons, these were selected for further studies to find out the role of the insect in the development of fruit rot.

3.2 Pathogenicity test

Healthy fruits of tomato cv. Pusa Ruby of the same age and size after being surface sterilized with 0.1 percent mercuric chloride solution and washed with sterile water were pin-pricked and subsequently inoculated by placing spore suspension of A. niger grown on potato-dextrose-agar. The inoculated specimens were transferred to sterilized desiccators over sterilized wire gauge. Pure culture of A. niger was obtained by hyphal-tip technique (Riker and Riker, 1936) and maintained on PDA slants.

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*Potato-dextrose-agar = Peeled potatoes 500 g (boiled+ exudates taken)
Dextrose = 20 g
Agar = 20g, Water to make 1000 ml.

3.3 Fungi associated with Drosophila busckii

Insects collected in above were studied for different fungi associated with them. These insects were dissected in the aseptic chamber and various parts separated with the help of sterilized needles. The parts so obtained were transferred to petridishes containing Potato-dextrose-agar. Fungi associated with insect body were also isolated by pouring the suspension on P.D.A. obtained by shaking different parts of the insects with 10 ml of sterile distilled water. The plates were incubated at 25°C. Fungi appearing were isolated and identified.

3.4 Inoculation of fruits with fungus

Throughout the studies the tomato fruits of the same size and age were inoculated with spores of the fungus grown on P.D.A. by pin-prick method. The spore suspension in sterile water was placed over the injured area of the fruit previously surface sterilized with 0.1 percent mercuric chloride.

3.5 Fruit surface fungi

Seedlings of tomato cv. Pusa Ruby were grown in fields. At the time of flowering and soon after fruit setting, they were bagged with butter paper to eliminate lodging of

further air-borne fungi. Unbagged fruits were kept for control. After a month of bagging the fungi were isolated by surface washing with sterile water. The fruits were washed with sterile distilled water and the washingswere transferred to petridishes containing P.D.A. The petridishes were incubated at 30°C. Fungi appearing were isolated and identified.

3.6 Rearing of insects

The insects were made to lay eggs on surface sterilized piece of vegetables in the laboratory under sterile conditions in glass jars. The adults developing were used for further studies. Throughout the studies, unless stated otherwise, the insects were allowed to feed on the fruits for 10, 20, 30 and 60 minutes under sterile conditions.

3.7 Observation and record of data

Throughout the studies the fruits after inoculation were kept in sterilized desiccators. This was followed by release of insects. Observations were made after 2, 5, 7, 10 and 15 days unless otherwise mentioned. The degree of rotting was noted as follows:

(-)	Nil	=	No infection
(+)	Poor	=	25% fruit surface infected
(++)	Moderate	=	25-50% fruit surface infected
(+++)	Severe	=	>50% fruit surface infected

Throughout the studies there were ten replicates for each treatment.

3.8 Effect of temperature on the development of fruit rot caused by A. niger in the presence of D. busckii

Tomato fruits were inoculated with the fungus and kept in air tight sterilized desiccators. Insects reared in the laboratory were released in the desiccator. The desiccators were transferred to cabinets maintained at 0°, 10°, 20° and 30°C. Fruits inoculated with fungus alone; fed with insect alone; and uninoculated with fungus and unfed with insects, served as control.

3.9 Effect of relative humidity on the development of fruit rot caused by A. niger in the presence of insects

Different relative humidities were maintained by aqueous supersaturated solutions of chemicals (Table,2). The solutions were kept in the lower portion of desiccators. Tomato fruits after having surface sterilized were inoculated with the fungus and transferred to the desiccators containing supersaturated solutions of chemicals. Later the insects were released. The desiccators were incubated at 20°C. Since a moderate infection was observed at 20°C, this temperature was selected for studying the effect of relative humidity on fruit rot.

Table 2. Chemicals used for maintaining different relative humidity (at 20°C)

Chemical	Relative humidity* (percent ± 1)
Sodium hydrogen sulphate ($NaHSO_4.H_2O$)	50
$NaBr.\ 2H_2O$	60
NH_4Cl and KNO_3	70
Ammonium chloride (NH_4Cl)	80
Potassium bromide (KBr)	85
Sodium hydrogen phosphate $Na_2HPO_4.\ 12H_2O$	95
Water (H_2O)	100

* Weast, R.C. and Astle, M.J. (1982)

3.10 Change in amino acid contents in tomato inoculated with A. niger and fed by insects

The fruits were inoculated with Aspergillus niger and fed by the insects Drosophila busckii for different intervals as outlined above. Fruits inoculated with fungus and those uninoculated and unfed with insect; and fed with insect alone, served as control. Qualitative analysis for amino acid was made by ascending paper chromatography. To 1 g of tomato tissues 10 ml of 70 percent ethanol was added. After 24 hrs these samples were macerated and centrifuged at 3000 r.p.m. for 15 minutes. The supernatant was dried at 60°C and redissolved in one ml of 70 percent ethanol. These samples were spotted on Whatmans filter paper no.1 by capillary tube and these strips were run with solvent n-butanol-acetic acid: water (4:1:5) v/v/v. The strips were dried and sprayed with 0.2 percent ninhydrin in water. Rf values so obtained were compared with the values of known amino acids for identification.

3.11 Ascorbic acid content in tomato fruits inoculated with A. niger and fed by D. busckii

The fruits inoculated with A. niger and fed with D. busckii separately and together were analysed for ascorbic acid along with unfed and healthy fruits. Ascorbic acid content was determined by titration method based on reduction of 2,6-dichlorophenol indophenol dye (Roe,1954).

3.11.1 Ascorbic acid standard solution

In order to prepare standard solution 50 mg of AR grade of ascorbic acid was dissolved in 50 ml of 5 percent HPO_3 solution and decanted in 200 ml of volumetric flask. The decanted solution was made upto the mark by adding required amount of 5 percent HPO_3. In this way 1 ml of this solution contained 0.2 mg of ascorbic acid. The solution was prepared fresh each time before use.

3.11.2 Preparation of 0.25% of 2,6-dichlorophenol indophenol reagent

The reagent was prepared by mixing 2,6-dichlorophenol indophenol with 150 ml of distilled water and warming gently until dissolved. To this 42 mg of $NaHCO_3$ was added. It was cooled and the volume was made to 250 ml in volumetric flask by adding required amount of distilled water.

3.11.3 Extraction from plant tissue

Dried tomato fruits (0.2 g) were throughly ground in 0.4 percent oxalic acid solution in water and centrifuged at 2,000 r.p.m. for 15 min. The supernatant was made upto 20 ml with oxalic acid solution. Five ml of this extract was titrated against standardized indophenol reagent. A pink colour indicated the end-point which however, persisted for only about fifteen seconds.

Ascorbic acid content of the plant extract was calculated by using following formula (Mahadevan and Sridhar's,1982)

$IxSx \frac{D}{A} x \frac{100}{W}$ = mg of ascorbic acid/100 g of tissues.

Where I = ml of indophenol reagent used in the titration.

S = mg of ascorbic acid reacting with one ml of the reagent.

D = Volume of the extract (in ml).

A = The aliquot titrated (in ml).

W = Dry weight of the sample (in g).

The data were subjected to statistical analysis.

3.12 Effect of treatment of fruits with ethanol on development of fruit rot in the presence of *D. busckii*

Healthy ripe tomato fruits were surface sterilized with ethanol (90 percent) and inoculated with the fungus and were kept in sterilized desiccators. Soon after inoculation specimens of *D. busckii* were released for different intervals i.e., 10, 20 and 30 minutes, 1, 6 and 24 hrs, 2 and 3 days separately. Fruits inoculated with fungus alone and those uninoculated and not fed by *D. busckii* served as control. There was another set of control without treatment with alcohol.

3.13 Effect of treatment of fruits with neem leaves on development of fruit rot

Leaves of neem (Azadirachta indica) were dried and ground to fine powder. The powder was passed through 100 mesh sieves and dusted on healthy moistened tomatoes. Freshly collected leaves of neem (20 g) were macerated in warring blendor in (100 ml) distilled water. The extract was passed through muslin cloth. Healthy ripe fruits were sprayed with this extract with atomiser. The fruits were inoculated with A. niger and fed with insect separately and together. The insects were allowed to feed for different intervals. Uninoculated and untreated fruits inoculated with fungus and fed with insect served as control.

3.14 Effect of leaf extracts of certain plants on development of fruit rot in presence of insect

Extracts of leaves of 19 different species of plants belonging to 14 families were prepared by macerating 10^1 g each of freshly collected thoroughly washed leaves in 100 ml of distilled water. The extract thus prepared was filtered through Whatman filter paper No.1. The filtrate was arbitrarily named "standard". Dilutions 0.01 and 0.001 were prepared by adding required amount of distilled water.(Table,3)

Saxena unpublished

Table 3. Plant species tested for the effect of leaf extract on fruit rot development

Family	Generic name of plant
Papaveraceae	Argemone mexicana Linn.
Caesalpinaceae	Cassia fistula Linn.
Solanaceae	Solanum xanthocarpum Schrad & Wendl.
	Withania somnifera (Linn.) Dunal
Verbenaceae	Lantana camara Linn.
Myrtaceae	Callistemon lanceolatus DC.
	Eucalyptus globulus Labill.
Labiatae	Mentha arvensis Linn.
	Ocimum sanctum Linn.
Asclepiadaceae	Calotropis procera (Ait.) R.Br.
Bignoniaceae	Adenocalymna alliacea (Lamk.) Miers
Meliaceae	Azadirachta indica Juss.
Euphorbiaceae	Euphorbia hirta Linn.
Chenopodiaceae	Chenopodium album Linn.
Umbelliferae	Peucedenum graveolans Hiern
	Foeniculum vulgare Mill.
Liliaceae	Allium cepa Linn.
	Allium sativum Linn.
Gramineae	Cymbopogon citratum (DC.) Staf.

Fruits were inoculated with fungus both prior and after treating with leaf extract. These were later fed by insects for different intervals. Appropriate controls with treated fruits fed with insect and inoculated with fungus alone separately; inoculated and unfed with insect both prior and after treatment; uninoculated with fungus and unfed with insect but treated with leaf extracts, were maintained.

3.15 Effect of growing tomato seedlings in soil amended with different oil cakes and bavistine on the development of fruit rot when inoculated with *A. niger* in presence of *D. buscki*

Soil (1 kg) was amended with oil cakes of castor (6 g), mustard (6 g), castor and mustard (3g + 3g), castor + bavistine (3g + 0.5g), mustard + bavistine (6g + 0.5g), castor + mustard + bavistine (6g + 6g + 0.5g) and bavistine only (0.5g). Seedlings of tomato grown in autoclaved soil were transplanted in these soils. Fruits so obtained were tested for the development of fruit rot both in presence and absence of insect. Fruits obtained from plants grown in unamended soil served as control.

CHAPTER IV

RESULTS

4.1 Survey of insects found in vegetable shops in Aligarh markets

Survey for various insects visiting the shops was made during rainy, summer and winter seasons. On these shops both household and plant pests were encountered almost in all the localities surveyed. Amongst the household pests, Musca domestica was very frequently seen visiting but their frequency ranged from 2-20 percent in different localities. However, the frequency was high during rainy season (table,4).

Frequency of Drosophila melanogaster, Ephestia cautella, Liposcelis divinatoria was low in all the localities and in some of the localities not even a single specimen was observed even in rainy season, a season most favourable for insect multiplication. The frequency of Drosophila busckii was highest in amongst different insects. When the three seasons were compared, the frequency was poor during hot season. It appears that moderate to low temperatures are very conducive for the multiplication of this insect. The frequency of other insects was moderate to low in all the localities and in all the three seasons.

Since, the frequency of D. busckii was very high in all the shops/localities where vegetables are sold this was selected for further studies.

Table 4. Frequency of occurrence of various insects in vegetable shops in ten different localities in Aligarh city

Name of insects	Order of insect	Frequency of insects in shops of different localities																											
		A			B			C			D			E			F			G			H			I			J		
		R*	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S
Musca domestica	Diptera	8**	2	20	20	8	15	10	3	20	18	10	16	18	6	10	20	6	15	18	9	10	18	9	10	20	6	15	20	6	9
Anopheles quadrimaculatus	Diptera	7	3	15	3	-	15	4	4	4	6	2	4	11	3	6	6	3	8	10	3	8	9	4	9	8	2	9	6	2	9
Culex fatigans	Diptera	9	-	8	9	3	8	9	4	6	6	5	5	9	4	8	8	5	9	5	4	7	5	5	4	6	-	8	9	-	3
Aedes aegypti	Diptera	11	-	7	6	5	9	5	-	7	6	-	6	6	-	4	9	2	4	2	-	2	3	2	3	4	1	-	5	-	5
Aedes albopictus	Diptera	9	2	8	4	6	7	6	-	8	8	-	10	3	-	3	4	1	6	5	-	7	3	2	6	5	-	7	8	3	8
Culex pipiens	Diptera	6	3	-	3	-	-	6	-	9	9	6	4	2	-	5	2	-	-	3	-	7	-	-	7	8	-	6	-	-	-
Drosophila busckii	Diptera	30	82	12	15	42	12	30	60	16	20	45	18	15	65	20	16	60	20	20	60	25	20	60	28	10	70	25	18	75	30
Drosophila melanogaster	Diptera	8	-	-	-	1	-	-	-	-	-	-	-	-	-	-	2	-	-	-	2	-	-	-	-	-	-	-	-	-	1
Ephestia cautella	Lepidoptera	-	-	-	2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	1	-
Liposcelis divinatoria	Psocoptera	-	2	-	-	-	-	-	4	4	-	9	6	15	9	2	4	2	-	-	-	2	2	-	-	-	-	1	-	-	1
Apis dorsata	Hymenoptera	-	2	9	15	10	15	10	8	12	10	4	7	12	7	12	8	8	11	15	6	9	15	6	12	16	9	10	12	6	12
Camponotus sp.	Hymenoptera	6	-	7	10	12	2	6	6	4	4	4	9	6	4	15	9	4	9	12	7	9	12	7	10	12	6	12	10	3	15
Polistes hebraeus	Hymenoptera	-	2	14	7	9	12	5	6	4	5	6	8	2	2	9	7	4	8	8	8	8	8	3	9	9	5	2	8	4	5
Coccinalids sp.	Coleoptera	6	2	-	6	4	5	9	5	6	8	9	7	1	-	6	5	5	10	2	1	6	5	2	2	2	1	4	4	-	2

*R = Rainy season = July; W = Winter = Dec/Jan; S = Summer = June

** Each figure is a mean of five samples spread over the whole period

A = Raghubirpuri; B = Sabzi mandi; C = Agra road; D = G.T. road; E = Vishnupuri; F = Dodhpur; G = Shamshad market; H = ITI road; I = Jaiganj; J = Barhaduari

4.2 Survey of fungi found associated with tomato fruits sold in the Aligarh market

It is clear from the results given (table,5) that in all thirteen fungi were found associated with rotting of tomato fruits in different localities, during the three seasons. The frequency of occurrence of Actinomucor sp, Alternaria tenuis, A. solani, Cladosporium fulvum, Curvularia sp, Geotrichum sp, and Fusarium sp. was low (below 10%) throughout the year in all the localities, whereas, the frequency of the remaining fungi was moderate to high (above 10%). Highest frequency of occurrence was observed for Aspergillus niger throughout the year in all the localities surveyed (table,5).

4.3 Pathogenicity of Aspergillus niger on tomato fruits

Although it has been shown earlier that A. niger is responsible for causing fruit rot of tomato, but in order to ascertain whether the isolate from Aligarh market is pathogenic in causing rot, pathogenicity tests were undertaken.

Tomato fruits inoculated with spores of A. niger, grown on potato-dextrose-agar, developed typical rotting symptoms in the form of soft water soaked areas with white mycelial growth later turning black. These symptoms were identical to those observed in fruits obtained from the market.

Table 5. Frequency of occurrence of various fungi found associated with rotting of tomato fruits in different markets of Aligarh

Fungi/localities	Frequency (%) of different fungi in three seasons																											
	A			B			C			D			E			F			G			H			I			J		
	*R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S	R	W	S
Actinomucor sp.	**-	-	-	-	-	-	-	-	-	-	-	2	-	1	1	-	-	-	-	-	2	-	-	1	-	-	-	-	-	1
Alternaria temuis	-	-	-	-	1	1	-	-	5	-	1	2	-	2	2	-	-	2	-	2	4	-	1	2	-	1	2	-	1	-
Alternaria solani	-	-	-	-	1	3	-	2	6	-	2	3	1	2	4	-	-	4	1	3	4	-	2	3	-	2	3	-	1	3
Cladosporium fulvum	-	-	-	-	2	5	-	1	3	-	1	2	-	1	2	-	1	5	-	3	2	-	1	2	-	-	3	-	2	2
Curvularia sp.	-	-	-	-	3	5	-	2	4	-	1	2	-	1	2	-	1	3	1	3	2	-	1	5	1	1	2	-	2	2
Geotrichum candidum	-	-	-	-	4	6	-	2	2	-	-	2	-	-	2	-	1	2	-	2	3	-	2	3	-	1	3	-	2	2
Fusarium sp.	-	-	-	-	4	5	-	2	2	-	2	4	-	2	2	-	2	2	-	4	2	1	2	2	-	2	3	1	-	3
Rhizopus arrhizus	10	10	17	10	10	8	10	10	11	10	9	10	11	8	12	10	10	12	10	8	10	10	10	7	10	12	11	10	10	11
Aspergillus niger	31	26	24	30	24	26	30	27	18	28	28	24	32	26	23	30	26	21	26	20	19	32	20	19	27	22	18	28	24	18
Rhizopus nigricans	22	20	17	20	13	14	20	17	14	20	17	17	17	19	13	20	19	17	20	18	13	17	19	18	20	19	17	20	18	16
Aspergillus flavus	15	20	15	20	15	14	20	17	15	20	18	10	20	18	15	20	20	12	20	17	17	20	20	18	20	17	15	20	16	15
Aspergillus fumigatus	10	11	15	10	11	7	10	10	10	10	11	10	10	10	12	10	10	10	10	10	11	10	11	10	12	13	10	11	12	13
Aspergillus nidulans	12	13	12	10	12	6	10	10	10	12	10	12	10	10	10	10	10	10	12	10	11	10	11	10	10	10	13	10	12	14

* R = Rainy season = July; W = Winter = Dec/Jan; S = Summer = June

** Each figure is a mean of five samples spread over the whole period

A = Raghubirpuri; B = Sabzi mandi; C = Agra road; D = G.T. Road; E = Vishnupuri; F = Dodhpur; G = Shamshad market; H = ITI road; I = Jaiganj; J = Barhaduari

It, therefore, reveals that the isolate of A. niger from the local market is pathogenic.

4.4 To detect the presence of inoculum on the body of insect

Since D. busckii was found commonly feeding on rotted fruits, it was considered desirable to find out if the insects carry spores of the fungus or not. Isolations were made directly from different parts of the body and also from washings of different parts during rainy season when rotting was highest. Results are presented in tables 6 and 7 . The inoculum of A. niger was found on all the parts of the insect body i.e., head, wings, abdomen and legs, collected from different localities, except, from legs of insect from Agra road and ITI road; wings and abdomen in Vishnupuri; and head and wings in Shamshad market. This could probably due to pose of sitting of the insect on infected fruits and the duration of sitting. A. flavus was isolated from all the parts of the body from Vishnupuri and Shamshad market. P. notatum was isolated from wings of insects from ITI road and head and wings of insects from Jaiganj market. Alternaria sp. was isolated from wings and abdomen of the insect from Raghubirpuri; legs of insects from Jaiganj and Barhaduari; Fusarium from wings, abdomen and legs of insects from Agra road, head of insects from Dodhpur, Abdomen and legs of insects from Barhaduari; R. stolonifer was isolated from abdomen and legs

Table 6. Fungi isolated from various parts of *Drosophila busckii* in different localities in Aligarh during rainy seasons

Fungi isolated/ localities	Fungi from different parts of insects body																			
	A				B				C				D				E			
	*H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L
Aspergillus niger	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	-	-	+
Aspergillus flavus	-	-	+	+	-	+	+	-	-	+	+	+	+	+	-	+	+	+	+	+
Penicillium notatum	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Alternaria sp.	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Fusarium sp.	-	-	-	-	-	-	-	-	-	+	+	+	-	-	-	-	-	-	-	-
Rhizopus stolonifer	-	-	+	+	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-	+

Fungi isolated/ localities	Fungi from different parts of insects body																			
	F				G				H				I				J			
	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L
Aspergillus niger	+	+	+	+	-	-	+	+	+	+	+	-	+	+	+	+	+	+	+	+
Aspergillus flavus	+	-	+	+	+	+	+	+	-	+	-	-	+	+	-	+	+	+	-	-
Penicillium notatum	-	-	-	-	-	-	-	-	-	+	-	-	+	+	-	-	-	-	-	-
Alternaria sp.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	+
Fusarium sp.	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+
Rhizopus stolonifer	+	-	+	+	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-

*H = Head; W = Wings; A = Abdomen; L = Legs; Fungus + (presence) ; - (absence)

A = Raghubirpuri; B = Sabzi mandi; C = Agra road; D = G.T.Road; E = Vishnupuri;
F = Dodhpur; G = Shamshadmarket; H = ITI road; I = Jaiganj; J = Barhaduari

Table 7. Fungi isolated from washings of various parts of *Drosophila busckii* from different localities of Aligarh during rainy season

Fungi isolated/ localities	A				B				C				D				E				F				G				H				I				J			
	*H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L	H	W	A	L
Aspergillus niger	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+
Aspergillus flavus	-	-	+	+	-	-	+	+	-	-	-	+	+	+	-	-	-	-	+	+	+	+	-	-	+	+	-	-	+	+	-	-	-	-	+	+	+	+	-	+
Penicillium notatum	-	-	-	+	+	+	-	-	+	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	-	-	-	+	-	-	-	-	-	-	+
Alternaria sp.	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	+	+	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	+	-	+
Fusarium sp.	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	+	+	+	-	-	-	-	+	+	+	-	+	-	-	+	-	-	-	+	-	-	-	+	-	-
Rhizopus stolonifer	-	-	+	+	-	-	-	-	-	-	+	+	-	+	-	-	+	-	-	+	-	+	+	-	+	-	-	-	+	+	-	-	+	+	-	-	-	-	+	+

* H = Head, W = Wings, A = Abdomen; L = legs; Fungus + (presence); - (absence)

A = Raghubirpuri; B = Sabzi mandi C = Agra road; D = G.T.road; E = Vishnupuri;
F = Dodhpur; G = Shamshad Market ; H = ITI road, I = Jaiganj;
J = Barhaduari

of insects from Raghubirpuri and ITI road, from head, abdomen and legs of insects from Dodhpur, abdomen of insects from Sabzi Mandi; and legs of insects from G.T. road and Vishnupuri. It is clear that inoculum of A. niger is borne on all the parts of insects in all the localities and thus appears to be an important carrier of A. niger. More or less identical results were obtained when isolations were made from washings from various parts of the insect body (table,7). The fact remains that all the parts of the insect body are capable of carrying inoculum of A. niger.

4.5 Effect of bagging of fruits in plants on fungi present on the fruits

It is evident from the results given in table 8 that when the fruits were bagged with butter paper, only one fungus (A. flavus) could find way to lodge on fruits while four fungi in unbagged. The number of fungi isolated from the fruits obtained from market was greater than those obtained from the standing crop. It was four as against 13 from the fruits obtained from the market. It appears that remaining fungi not isolated here came in association with the fruit during transport and storage or when the fruits over ripened they came as saprophytes.

Table 8. Fungi isolated from bagged and unbagged tomato fruits

Fungi isolated	Bagged fruits	Unbagged fruits
Aspergillus niger	-	+
Aspergillus flavus	+	+
Alternaria pori	-	+
Rhizopus sp.	-	+

There were five plants for each treatment which was replicated thrice

- (absent)
+ (present)

4.6 Effect of temperature on the development of fruit rot of tomato caused by A.niger in the presence of D.busckii

There has been no rotting in fruits upto 15 days at 0°C in all the treatments. However, at 10°C and 20°C rotting was observed at 15 days in all except in uninoculated ones. There was slight softening of fruits at these temperatures in uninoculated,which could be due to natural senescence. At 30°C not only rotting occurred earlier but intensity was also high. Thus high temperatures are conducive for rotting, and in the presence of insect the rotting appeared earlier.

In the fruits fed with insect there has been some rotting due to the fungus which might be due to spores escaping through insect body. But at 30°C the rotting was observed after 10 days in fungus inoculated and 7 days in fungus inoculated and insect fed fruits. Therefore, the highest rotting appeared at 30°C in the present studies (table,9).

4.7 Effect of relative humidity on development of fruit rot caused by A. niger in the presence of D. busckii

From the table,10 it is clear that at low R.H. i.e., 50, 60 and 70 percent initiation of rotting occurred after 7 days, while at 80 and 85 percent after 5 days and at 95 and 100 percent after 2 days. However, at 95 and 100 percent severe rotting was observed after 5 and 7 days respectively. At the remaining relative humidity 7 or more days were

Table 9. Effect of temperature on rotting of tomato fruits due to Aspergillus niger in the presence of Drosophila busckii

Treatments	Temp. (°C)	Rotting after (days)				
		2	5	7	10	15
Uninoculated	0	-	-	-	-	-
	10	-	-	-	-	-
	20	-	-	-	-	-
	30	-	-	-	-	++
Fungus	0	-	-	-	-	-
	10	-	-	-	-	++
	20	-	-	-	-	++
	30	-	-	-	++	+++
Insect fed and inoculated with fungus	0	-	-	-	-	-
	10	-	-	-	-	+++
	20	-	-	-	-	+++
	30	-	-	+	++	+++
Insect fed alone	0	-	-	-	-	-
	10	-	-	-	-	+
	20	-	-	-	-	+
	30	-	-	-	-	++

(-) Nil, (+) Poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates.

Table: Effect of temperature on rotting of tomato fruits due to Aspergillus niger in the presence of Drosophila buscki

Treatments	Temp. (°C)	Rotting after (days) 2	5	7	10	15
Uninoculated	0	-	-	-	-	0
	10	-	-	-	-	0
	20	-	-	-	-	0
	30	-	-	-	-	2.1 b
Fungus	0	-	-	-	-	-
	10	-	-	-	-	2.2 b
	20	-	-	-	-	2.1 b
	30	-	-	-	2.0 b	3.0 c
Insect fed and inoculated with fungus	0	-	-	-	-	-
	10	-	-	-	-	3.0 c
	20	-	-	-	-	3.1 c
	30	-	-	1.1 a	2.1 b	3.1 c
Insect fed alone	0	-	-	-	-	-
	10	-	-	-	-	1.0 a
	20	-	-	-	-	1.1 a
	30	-	-	-	-	2.1 b

(-) Nil = No infection, (1) Poor = 25% fruit surface infected,
(2) Moderate = 25-50% fruit surface infected and
(3)-Severe = >50% fruit surface infected.
Means in the same column followed by different letters are significantly different at P = 0.05.

Fig.2.1 Effect of different temperatures on the development of fruit rot caused by A.niger in the presence of D. busckii

0°C

CONTROL INSECT INSECT-FUNGUS FUNGUS

10°C

CONTROL INSECT INSECT-FUNGUS FUNGUS

Fig.2.2 Effect of different temperatures on the development of fruit rot caused by A.niger in the presence of D. busckii

20°C

30°C

Table 10. Effect of different relative humidity (R.H.) on the development of fruit rot of tomato caused by Aspergillus niger in presence of Drosophila busckii

Relative Humidity (%)	Treatments (Inoculated with or fed with)	Rotting after (days)				
		2	5	7	10	15
50	Uninoculated	-	-	-	++	+++
	Fungus	-	-	++	+++	+++
	Insect + fungus	-	-	-	+++	+++
	Insect	-	-	-	++	+++
60	Uninoculated	-	-	-	++	+++
	Fungus	-	-	++	+++	+++
	Insect + fungus	-	-	-	+++	+++
	Insect	-	-	-	++	+++
70	Uninoculated	-	-	-	++	+++
	Fungus	-	-	++	+++	+++
	Insect + fungus	-	-	-	+++	+++
	Insect	-	-	-	++	+++
80	Uninoculated	-	-	+	++	+++
	Fungus	-	+	++	+++	+++
	Insect + fungus	-	+	++	++	++
	Insect	-	-	++	++	+++
85	Uninoculated	-	-	++	+++	+++
	Fungus	-	+	++	+++	+++
	Insect + fungus	-	+	++	+++	+++
	Insect	-	-	++	+++	+++
95	Uninoculated	-	+	++	+++	+++
	Fungus	+	+	+++	+++	+++
	Insect + fungus	+	++	+++	+++	+++
	Insect	-	++	+++	+++	+++
100	Uninoculated	-	++	+++	+++	+++
	Fungus	+	+++	+++	+++	+++
	Insect + fungus	+	+++	+++	+++	+++
	Insect	-	+++	+++	+++	+++

(-) Nil, (+) poor, (++) moderate and (+++) severe
Each value is mean of 3 replicates.

Table: Effect of different relative humidity (R.H.) on the development of fruit rot of tomato caused by Aspergillus niger in presence of Drosophila busckii.

Relative humidity (%)	Treatments (Inoculated with or fed with)	Rotting after (days)				
		2	5	7	10	15
50	Uninoculated	-	-	-	2.1 b	3.1 c
	Fungus	-	-	2.1 b	3.1 b	3.2 c
	Insect+Fungus	-	-	-	3.2 c	3.2 c
	Insect	-	-	-	2.2 b	3.1 c
60	Uninoculated	-	-	-	2.0 b	3.1 c
	Fungus	-	-	2.0 b	3.1 c	3.1 c
	Insect+Fungus	-	-	-	3.2 c	3.2 c
	Insect	-	-	-	2.1 b	3.2 c
70	Uninoculated	-	-	-	2.0 b	3.3 c
	Fungus	-	-	2.0 b	3.1 c	3.1 c
	Insect+Fungus	-	-	-	3.0 c	3.2 c
	Insect	-	-	-	2.0 b	3.2 c
80	Uninoculated	-	-	1.0 a	2.0 b	3.1 c
	Fungus	-	1.0 a	2.1 b	3.1 c	3.1 c
	Insect+Fungus	-	1.1 a	2.2 b	2.2 b	2.1 b
	Insect	-	-	2.1 b	2.1 b	3.2 c
85	Uninoculated	-	-	2.1 b	3.1 c	3.2 c
	Fungus	-	1.0 a	2.0 b	3.2 c	3.2 c
	Insect+Fungus	-	1.1 a	2.2 b	3.2 c	3.1 c
	Insect	-	-	2.2 b	3.3 c	3.1 c
95	Uninoculated	-	1.2 a	2.3 b	3.2 c	3.2 c
	Fungus	1.1 a	1.2 a	3.0 c	3.1 c	3.2 c
	Insect+Fungus	1.1 a	1.2 a	3.1 c	3.1 c	3.2 c
	Insect	-	1.2 a	3.0 c	3.0 c	3.2 c
100	Uninoculated	-	1.2 a	3.0 c	3.0 c	3.2 c
	Fungus	1.1 a	3.0 c	3.1 c	3.0 c	3.3 c
	Insect+Fungus	1.2 c	3.1 c	3.1 c	3.1 c	3.1 c
	Insect	-	3.2 c	3.2 c	3.1 c	3.1 c

(-) Nil = No infection, (1) Poor = 25% fruit surface infected,
(2) Moderate = 25-50% fruit surface infected and
(3)- Severe = >50% fruit surface infected.
(4) Means in the same column followed by different letters are significantly different at P = 0.05.

required for severe rotting to develop. Therefore, high relative humidity is very conducive for rotting and moreover, the rate of rotting is also influenced by the high humidity. In amongst various treatments the initiation of rotting was early when inoculated with fungus alone at 50 and with fungus alone and insect + fungus at the remaining relative humidity.

For studying the effect of relative humidity on development of fruit rot relative humidity above the average relative humidity of Aligarh (i.e., above 50 percent) was taken as the base.

4.8 Changes in ascorbic acid (Vitamin C) content in tomato fruits inoculated with *A. niger* and fed by *D. busckii*

The ascorbic acid content decreased with the increase in incubation period in all the treatments. But the decrease in the ascorbic acid was highest in fruits inoculated with *A. niger* alone where it was 86.3 percent after 10 days of incubation. In those inoculated with insect alone it was 39.1 and increased as the feeding duration increased. However, when fruits were inoculated with fungus and fed with insect the reduction was more than insect alone but less than fungus alone. Here also the reduction increased with the increase in the feeding duration of the insect. The reduction in the

Table 11. Changes in ascorbic acid content in tomato fruits inoculated with *Aspergillus niger* and fed with *Drosophila busckii* separately and together

Treatments	Duration of insect feeding (min)	Ascorbic acid content in rotted fruits (mg/100 g) ** Degree of rotting 0 AC	0 R	2 AC	2 R	4 AC	4 R	6 AC	6 R	10 AC	10 R	Loss of ascorbic acid after 10 days of incubation (per cent)
Uninoculated with fungus and unfed with insect	-	46*	-	44	-	37	+	34	+	33	++	28.2
	10	46	-	40	+	38	++	32	++	28	++	39.1
Insect	20	46	-	38	+	36	++	30	++	20	++	56.6
	30	46	-	32	+	30	++	20	++	14	+++	69.5
	10	44	-	40	+	37	++	32	++	12	+++	72.7
Insect + fungus	20	44	-	36	+	31	++	18	+++	10	+++	77.2
	30	44	-	30	+	20	++	12	+++	8	+++	81.8
Fungus	-	44	-	32	+	24	++	16	+++	6	+++	86.3
L.S.D. at 1% level		2.5		1.8		2.9		3.0		3.1		3.5
L.S.D. at 5% level		1.9		1.3		1.8		1.5		1.5		1.9

* Ascorbic acid content, ** Degree of rotting, AC = Ascorbic acid, R = Rotting

ascorbic acid content in fruits inoculated with A. niger and fed with insects for 30 min was almost the same as that of fungus alone after 10 days of inoculation (table,11).

4.9 Changes in amino acid content in tomato fruits inoculated with A. niger and fed by D. busckii

It is evident that eleven amino acids were detected when fed with insect alone; eleven in insect and fungus, and five with fungus alone as compared to seven in healthy tomatoes. The number of amino acids detected after different intervals varied. Arginine was detected in healthy as well as in those fed with the insect while; isoleucine, alanine, valine and asparagine were detected in fruits fed by insect and inoculated with fungus and methionine was absent in those inoculated with fungus alone; while amino acetic acid, glutamic acid and tryptophane were absent in those fruits fed by insect alone. Histidine, tyrosine, lysine, cysteine and isoleucine were present in those inoculated with fungus alone (table,12).

4.10 Studies on control of fruit rot caused by A. niger in presence of D. busckii

4.10.1 Effect of treatment with ethanol.

When tomatoes were inoculated with A. niger and fed with Drosophila busckii for different intervals,rotting was very high in the presence of insect. It increased with the

Table 12. Amino acids detected in tomato fruits inoculated with Aspergillus niger and fed with Drosophila busckii separately and together

Amino acids	Fed with insect				Inoculated with fungus and fed with insect				Inoculated with fungus alone				Healthy
	Rotting after days				Rotting after days				Rotting after days				
	2	4	6	10	2	4	6	10	2	4	6	10	•
Histidine	$+^{a}+^{c}$	$+^{c}$	$+^{b}$	$+^{b}$	-	$+^{a}$	$+^{a}$	-	+	+	-	+	+
Tryrosine	$+^{a}+^{c}$	-	$+^{b}+^{a}$	$+^{b}$	$+^{b}+^{c}$	$+^{c}$	$+^{a}$	$+^{a}+^{c}$	+	+	-	+	-
Lysine	$+^{a}+^{b}$	$+^{a}$	$+^{a}+^{c}$	-	$+^{a}+^{c}$	$+^{c}$	$+^{c}$	-	-	-	+	-	+
Arginine	-	-	-	$+^{c}$	-	-	-	-	-	-	-	-	+
Tryptophane	-	-	-	-	-	$+^{b}$	-	-	-	-	-	-	+
Aspartic acid	-	$+^{b}$	-	$+^{b}$	-	-	-	$+^{b}$	-	-	-	-	+
Cysteine	-	$+^{a}$	-	$+^{a}+^{c}$	-	-	-	-	-	-	+	-	+
Methionine	-	$+^{c}$	-	$+^{b}$	-	-	-	-	-	-	-	-	+
Isoleucine	-	-	-	$+^{c}$	$+^{a}+^{b}+^{c}$	$+^{c}$	$+^{a}+^{b}$	$+^{a}$	-	-	+	+	-
Alanine	$+^{a}$	$+^{b}$	-	-	-	-	-	$+^{a}+^{c}$	-	-	-	-	-
Valine	-	-	$+^{a}$	-	$+^{a}+^{c}$	-	$+^{b}$	$+^{b}$	-	-	-	-	-
Glutamic acid	-	-	-	-	-	$+^{a}$	-	-	-	-	-	-	-
Asparagine	-	-	$+^{c}$	$+^{a}$	-	$+^{b}$	-	-	-	-	-	-	-
Amino acetic acid	-	-	-	-	-	$+^{c}$	$+^{c}$	-	-	-	-	-	-

a = 10 min + present

b = 20 min - absent

c = 30 min

Each value is mean of 3 replicates

increase in duration of insect feeding, upto 30 minutes but prolonged feeding resulted in the slight decrease. The appearance of rotting was earlier when fed with D. busckii for 30 minutes i.e., rotting appeared after 2 days of insect feeding. However, in low period of feeding and prolonged feeding the rotting was delayed. It therefore, appears that insect brought about earlier appearance of rotting. It is understandable that insect through its saliva may have provided stimulus for fungus to grow on the fruit. The fruits treated with alcohol remained free of infection for short duration probably because of volatile nature of alcohol. But this preliminary study shows that alcohol could be tried in plant extract for controlling of the fruit rot (table,13). Therefore, in the following studies alcoholic extract of certain plant species have been tested.

4.10.2 Ethanolic leaf extract of Lantana camara, Mentha arvensis and Ocimum sanctum

When fruits were treated with ethanolic leaf extract of L. camara, M. arvensis and O. sanctum, the fruits remained free from rotting upto 10 days in uninoculated with fungus and unfed with insect. Almost identical results were obtained when fruits were treated with ethanolic extract of L. camara prior to and after inoculation with fungus. The rotting was observed after 2 days when insects were allowed to feed for

Table 13. Effect of treating the fruits with alcohol on development of fruit rot caused by Aspergillus niger in the presence of Drosophila buscki for different durations

Insect duration		Rotting of fruits after (days)				
		2	5	7	10	15
10 min	a	-	+	+	++	+++
	b	+	+	++	+++	+++
20 min	a	-	+	++	++	++
	b	+	+	++	+++	+++
30 min	a	+	++	++	++	+++
	b	++	+++	+++	+++	+++
1 hr	a	-	+	+	++	++
	b	+	++	++	++	+++
6 hr	a	-	+	++	++	+++
	b	+	++	++	++	+++
24 hr	a	-	+	+	+	++
	b	+	++	++	++	+++
2 days	a	-	+	+	++	++
	b	+	++	++	+++	+++
3 days	a	-	+	+	++	++
	b	+	++	++	++	+++
Without insect (Fungus alone)	a	-	-	+	++	+++
	b	+	+	+	++	+++

(a) alcohol sterilized

(b) not sterilized with alcohol

(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates.

10, 20 and 30 minutes, in inoculation with fungus both prior and after the treatment. The rotting was less when fungal inoculation was done after the treatment with ethanolic extract. Fruits remained free from rotting upto 5 days when fed with insect for 10 minutes and upto 15 days when fed with insect for 20 and 30 minutes as a result of treatment with extract.

When fruits were treated with ethanolic extract of M. arvensis the rotting took place after 5 days in uninoculated and unfed. Similar resultswere obtained in fruits treated prior to and after inoculation. The rotting was seen after 7 days when inoculation with fungus was done after treatment followed by feeding by insect for 10, 20 and 30 minutes but in corresponding feeding insect duration with treatment prior to fungal inoculation the rotting started after 2 days. Fruits remained free from rotting upto 10 days when treated fruits were fed with insect only, in all feeding duration.

Similarly, when fruits were treated with ethanolic extract of O. sanctum the fruits remained free from rotting upto 10 days in uninoculated and unfed with insect. Rotting took place earlier i.e., after 2 days when fungus inoculation was done after treatment and after 5 days prior to treatment. Fruits remained free from rotting upto 7 days when inoculation was done after treatment and fed with insect for 10, 20 and 30 minutes. The rotting was less in fungal inoculation

Table 14. Effect of treatment of fruits with ethanolic extract of leaves of Lantana camara, Mentha arvensis and Ocimum sanctum on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

Treatments		Rotting of fruits after (days) L.camara 2	5	7	10	15	M.arvensis 2	5	7	10	15	O. sanctum 2	5	7	10	15
Uninoculated and unfed	a*	-**	-	-	-	+	-	-	+	+	++	-	-	-	-	++
	b	-	-	+	++	+++	-	-	+	++	+++	-	-	+	++	++
Inoculated with fungus after treatment	a	-	-	+	++	+++	-	++	++	++	++	-	+	+	++	++
	b	+	+	+	++	+++	+	+	+	++	+++	+	+	+	++	++
Inoculated with fungus before treatment	a	-	-	+	++	+++	-	++	++	++	+++	-	-	+	++	++
	b	+	+	+	++	+++	+	+	+	++	+++	+	+	+	++	++
Inoculated with fungus and insect fed (mts) after treatment																
10	a	-	+	+	+	+++	-	-	-	++	++	-	-	-	+	+
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
20	a	-	+	+	+	++	-	-	-	++	++	-	-	-	+	++
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
30	a	-	+	+	++	++	-	-	-	++	+++	-	-	-	++	+++
	b	++	+++	+++	+++	+++	++	+++	+++	+++	+++	++	+++	+++	+++	+++
Inoculated with fungus and insect fed (mts) before treatment																
10	a	-	+	+	+	+++	-	++	++	++	+++	-	-	++	+++	+++
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
20	a	-	+	+	+	+++	-	++	++	+++	+++	-	-	-	-	++
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
30	a	-	+	+	++	+++	-	++	++	+++	+++	-	-	-	-	++
	b	++	+++	+++	+++	+++	++	+++	+++	+++	+++	++	+++	+++	+++	+++
Insect fed alone (mts)																
10	a	-	-	++	++	++	-	-	-	-	+	-	-	-	++	++
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
20	a	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
	b	+	+	++	+++	+++	+	+	++	+++	+++	+	+	++	+++	+++
30	a	-	-	-	-	-	-	-	-	++	++	-	-	-	-	-
	b	++	+++	+++	+++	+++	++	+++	+++	+++	+++	++	+++	+++	+++	+++

*a = Treated with ethanolic extract
b = Not sterilized with ethanol
**(-) Nil, (+) poor, (++) moderate and (+++) severe
Each value is mean of 3 replicates

done after the treatment with extract. No rotting was observed upto 7 days when treated fruits were fed with insect only for 10 minutes but treated fruits remained completely free from rotting upto 15 days when fed with insect for 20 and 30 minutes.

Thus, ethanolic extracts of the leaf of L. camara and O. sanctum proved to be more effective than M. arvensis in controlling the fruit rot caused by fungi, both in the presence or absence of insect (table, 14).

4.10.3 Dry powder of leaves and extract of leaves of neem in water and alcohol

Results presented in table, 15 show that there has been no rotting of fruits upto 15 days in uninoculated and unfed fruits, when treated with neem leaf dry powder. The rotting of fruits treated with neem leaf powder before fungal inoculation was observed after 10 days. But on the contrary, in the fruits treated with neem leaf powder after fungal inoculation the rotting developed after 7 days. In the presence of insect the rotting developed after 10 days when fungus inoculation was done after treatment, with 10 minutes of the presence of insect and after 7 days with 20 minutes/30 minutes of the presence of insects. However, in the presence of insect with fungus inoculation before treatment, the onset of rotting was earlier. When fruits were fed with insect

alone no rotting occurred upto 10 days with different duration of insect feeding.

Time required for (++) type of rotting was more than 15 days in uninoculated unfed; fungus inoculated both before and after treatment; 7-10 days with insect with fungus inoculation after treatment, 7 days with insect with fungus before treatment and 15 days with insect alone.

When the results obtained with leaf powder were compared with those of water and alcoholic extract of leaf there was less rotting in the leaf extract (of both types) in fungus inoculated fruits as compared to those treated with dry powder. There was hardly any difference in the degree of rotting when inoculated with fungus after treatment in the presence of insect in the three types of leaf preparations. Rotting was almost identical in dry powder and leaf extract in water when fruits were inoculated with fungus in the presence of insect before treatment, However, in ethanolic extract of leaf there was early initiation of rotting in fungus inoculated before treatment in the presence of insect for 10 minutes. With 20 and 30 minutes of feeding there was no difference in three types of leaf preparations. The leaf extracts both alcoholic and water were effective in minimising the rotting when fed with insect alone as compared to leaf powder.

e 15. Effect of treatment of fruits with leaf of Azadirachta indica on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

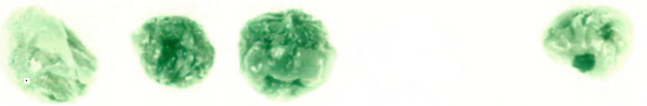
Treatments		Rotting of fruits after (days)													
		Dry powder					Leaf extract (water)					Leaf extract (ethanol)			
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
oculated and unfed	a*	-**	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	-	+	+++	+++	-	-	+	++	+++	-	-	+	+++
ulated with fungus	a	-	-	-	++	++	-	-	+	+	+	-	-	+	+
r treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus	a	-	-	++	++	++	-	-	+	+	+	-	-	+	+
re treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus and ct fed (mts) r treatment															
10	a	-	-	-	++	+++	-	-	+	++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	++	++	+++	-	-	++	++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	++	+++	+++	-	-	++	+++	+++	-	-	++	+++
	b	-	+	++	+++	++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus insect fed (mts) re treatment															
10	a	-	-	++	+++	+++	-	-	++	+++	+++	+	+	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ct fed alone (mts)															
10	a	-	-	-	-	+	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	+	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	+	-	-	-	-	-	-	-	-	-
	b	-	+	+	+	+	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	++	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	+++	-	+	+	+++	++	-	+	+	+++

*a = Treated with leaf extract
b = Control (untreated)
**(-) Nil, (+) Poor, (++) moderate and (+++) severe
Each value is mean of 3 replicates

Fig.3 Effect of leaf extract and dry powder of neem on the development of fruit rot caused by *A. niger* in the presence of *D. busckii*

TOMATO

NEEM EXTRACT CONTROL



10 20 30

(min)

TOMATO

NEEM DRY POWDER CONTROL

10 20 30

(Min.)

4.10.4 Effect of water extracts of certain plants on the development of fruit rot caused by A. niger in the presence of D. busckii

4.10.4.1 Argemone mexicana

Results presented in table, 16 show that no rotting was observed upto 7 days in those uninoculated with fungus and unfed with insect in 'S' and 1% dilution but rotting started after 5 days followed by moderate to severe in 0.1% conc. When fruits were treated with 'S', 1% and 0.1% there has been slight difference in treatment with leaf extract both prior and after fungal inoculation. In both the rotting was seen after 5 - 7 days in treated after inoculation and insect fed for 10, 20 and 30 minutes in all the concentrations but when inoculation was done prior to treatment the rotting was observed after 5 days in 'S' conc. and after 2 days in 1% and 0.1%. The rotting was less when fungal inoculation was done after the treatment with extract. The efficacy of the leaf extract in minimising the rotting decreased with the increase in dilution of the extract.

Time required for the development of rotting to the extent of ± 50 percent fruit cover (++) was 7-10 days in 'S'; and 5-7 days in 1% and 0.1%.

16. Effect of treatment of fruits with different concentrations of the leaf extract of Argemone mexicana on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

reatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	1
culated and unfed	a*	-**	-	-	++	++	-	-	-	++	++	-	-	++	++	+
	b	-	-	+	+++	+++	-	-	+	++	++	-	-	+	+++	+
lated with fungus treatment	a	-	-	+	++	+++	-	-	+	++	+++	-	+	+	+++	+
	b	-	+	++	++	+++	-	+	++	+++	+++	-	+	++	+++	+
lated with fungus e treatment	a	-	-	++	++	+++	-	-	++	++	++	-	+	++	+++	+
	b	-	+	++	+++	+++	-	+	++	++	++	-	+	++	+++	+
lated with fungus and t fed (mts) after ment																
10	a	-	-	-	++	+++	-	-	++	++	+++	-	-	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
20	a	-	-	++	++	+++	-	-	++	++	+++	-	-	++	+++	+
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	+++	+
30	a	-	-	+	++	+++	-	-	++	+++	+++	-	-	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
lated with fungus nsect fed (mts) e treatment																
10	a	-	-	++	++	++	-	++	++	++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
20	a	-	-	++	++	++	-	++	++	+++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
30	a	-	-	++	++	++	-	+	++	++	++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
t fed alone (mts)																
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	++	+
	b	-	+	+	+	++	-	+	+	++	++	-	+	+	++	+
20	a	-	-	-	-	++	-	-	-	-	++	-	-	++	++	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	+
30	a	-	-	-	+	++	-	-	-	-	++	-	-	++	++	+
	b	-	+	+	+++	+++	-	+	+	+++	+++	-	+	++	+++	+

*a = Treated with leaf extract
b = Control (untreated)
** (-) Nil, (+) Poor, (++) moderate and (+++) severe
Each value is mean of 3 replicates

4.10.4.2 Cassia fistula

Rotting of fruits when treated with 'S' concentration of leaf extract was observed in treated fruits uninoculated and unfed with insects after 7th day. There has been no difference in the rotting of fruits when inoculated with fungus before and after treatment. Similarly, there has been no difference in the rotting of fruits in the presence of insects for different duration in treatments before and after fungal inoculation except in prolonged duration of insect feeding (for 30 minutes) before treatment where the onset of rotting was earlier. In insect fed alone for 10 minutes the fruits remained free of rotting upto 15 days and 10 days with prolonged duration (i.e.,20 and 30 minutes). Dilutions of the extract reduced the efficacy of the extract in minimising the rotting.

Time required for the development of rotting to the extent of (++) was 7 - 10 days in uninoculated and unfed, 5-7 days in inoculated with fungus before and after treatment; and inoculation of fungus in presence of insects for different duration before and after treatment in different concentration of leaf extract. However, in insect fed alone it was 10 days or more than 15 days in different concentrations of the extract (table,17).

le 17. Effect of treatment of fruits with different concentrations of the leaf extract of Cassia fistula on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
noculated and unfed	a*	-**	-	+	++	+++	-	-	++	++	+++	-	-	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
culated with fungus	a	-	+	++	++	+++	-	-	++	++	+++	-	++	++	++
er treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus	a	-	+	++	++	+++	-	-	++	++	+++	-	++	++	++
ore treatment	b	-	+	++	+++	+++	-	+	++	+++	++	-	+	++	+++
culated with fungus insect fed (mts) er treatment															
10	a	-	-	++	++	+++	-	-	++	++	+++	-	+	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	++	++	+++	-	-	++	++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	++	+++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus insect fed (mts) ore treatment															
10	a	-	-	++	++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	++	++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	++	++	++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ect fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	+	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	+	-	-	-	-	++	-	-	-	-
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	+	-	-	-	-	-	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil , (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

4.10.4.3 Solanum xanthocarpum

There has been no rotting in 'S' concentration of the leaf extract of Solanum xanthocarpum upto 15 days and in 1% and 0.1% upto 7 days in uninoculated and unfed fruits. However, in inoculated with fungus before and after treatment with 'S' concentration of leaf extract the rotting started from 7th and 10th day respectively. Thus the fruit remained free for longer duration when treated fruits were inoculated.

In the presence of insects the period for which the fruits could be kept free from rotting was reduced to 5 days when insects were there for 20 and 30 minutes and 7 days for 10 minutes. The degree of rotting was more in both inoculated before and after treatment. The rotting although started on 5th day but its incidence was low in inoculated with fungus in the presence of insects for 30 minutes before treatments.

When fed with insect alone for 10 minutes there has been no rotting upto 15 days but rotting was observed on 10th and 15th day when duration of feeding was increased to 20 and 30 minutes respectively.

Time required for the development of rotting to the extent of 50 percent fruit cover (++) was 10 and 15 days respectively in fungus inoculation after treatment in 'S' conc.(table,18).

e 18. Effect of treatment of fruits with different concentrations of the leaf extract of *Solanum xanthocarpum* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila busckii*

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
oculated and unfed	a*	-**	-	-	-	-	-	-	-	++	+++	-	-	-	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
ulated with fungus r treatment	a	-	-	-	++	+++	-	-	++	+++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus re treatment	a	-	-	+	+	++	-	-	+	++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus insect fed (mts) r treatment															
10	a	-	-	++	++	+++	-	+	++	++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	+	++	++	+++	-	+	++	++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	+	++	++	++	-	-	++	++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ulated with fungus insect fed (mts) re treatment															
10	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	++
	b	-	+	+++	++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	+	+	+	+	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ct fed alone (mts)															
10	a	-	-	-	-	-	-	-	+	+	++	-	-	+	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	+	++	-	-	-	+	++	-	-	-	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	+	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	+++	-	+	++	++	++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

4.10.4.4 Withania somnifera

The rotting of fruits due to A. niger was initiated after 7 days when treated with the 'S' conc. of extract before and after inoculation; 7 days in inoculated with fungus after treatment in the presence of insects for 10 and 20 minutes; and for 30 minutes it started after 5 days. On the other hand, in the presence of insects and inoculated with fungus before the treatment the rotting started after 5 days irrespective of the duration of the presence of insects. Thus, treatment before inoculation proved better than after inoculation. In fruits fed with insect alone, there has been no rotting for 15 days. Increased dilutions brought about decrease in the efficacy of the extract.

Time required for the development of (++) type of rotting was 7 days in fungus inoculated alone and fungus inoculated in the presence of insect for 10 minutes; 5-10 days in the presence of insect for 20 minutes and 7-10 days in the presence of insects for 30 minutes when treated before inoculation in different concentration of the extract but when treated after inoculation the time required ranged from 5-7 days and in insect fed alone 10-15 days as against 10 days in uninoculated and unfed in all the concentrations of extracts. Thus more rotting was observed when fruits were treated with extract after inoculation in the presence of insect (table,19).

le 19. Effect of treatment of fruits with different concentrations of the leaf extract of Withania somnifera on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
noculated and unfed	a*	-**	-	-	++	+++	-	-	-	++	++	-	-	+	++
	b	-	-	+	++	+++	-	-	+	++	+++	-	-	+	+++
culated with fungus	a	-	-	++	++	++	-	-	++	++	++	-	+	++	+++
er treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus	a	-	-	++	++	+++	-	-	++	++	+++	-	+	++	++
ore treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus insect fed (mts)															
er treatment 10	a	-	-	++	++	++	-	+	++	+++	+++	-	+	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	++	++	-	-	++	++	++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	+	+	++	+++	-	+	++	++	++	-	+	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus insect fed (mts)															
ore treatment 10	a	-	+	++	++	+++	-	+	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	+	++	++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	+	++	++	+++	-	++	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ect fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	+	++	-	-	-	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	++	-	-	-	+	++	-	-	-	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+++	+++	++++	-	+	++	+++	++++	-	+	++	++++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

4.10.4.5 Lantana camara

Flower extract - Results presented in table,20 show that fruits treated with standard extract of flower of L. camara remained almost free from rot for 7-15 days both in uninoculated with fungus and unfed with insect and those fed with insect alone. When treatment with flower extract was given both prior and after fungal inoculation the rotting was initiated after 10 days but the degree of rotting was less when inoculation was done after treatment. When insects were allowed to feed for 10, 20 and 30 minutes, the rotting was less when inoculation was done after treatment.

With increase in dilution the protection period decreased. It was 5-7 days in 1% and 2-5 days in 0.1% in infected fruits. However, in uninoculated unfed and insect fed alone the degree of rotting was less (poor) after 10 and 15 days but in others it was moderate to severe after these periods.

Time required to develop rotting to the extent of 50 percent fruit cover (++) was 10 days in 'S'; and 7-10 days in 1% and 0.1%. When different treatments were compared with 'S' it was >15 days in uninoculated and unfed; and insect alone; 10 days in fungal inoculated prior treatment with extract; 15 days in fungal inoculation after treatment; and 10 days when insect were allowed to feed both prior and after treatment; in 1% >15 days in uninoculated and unfed and fed with

20. Effect of treatment of fruits with different concentrations of the flower extract of Lantana camara on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
oculated and unfed	a*	-**	-	-	+	+	-	-	-	+	+	-	-	-	+
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
lated with fungus treatment	a	-	-	-	++	++	-	-	+	++	+++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus re treatment	a	-	-	-	++	+++	-	-	-	++	+++	-	-	-	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) treatment															
10	a	-	-	-	+	+	-	-	+	++	++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	-	++	++	-	-	-	++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	-	++	++	-	-	-	++	+++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) re treatment															
10	a	-	-	-	++	+++	-	-	-	++	++	-	-	+	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	-	++	+++	-	-	-	++	++	-	-	+	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	-	++	+++	-	-	++	+++	+++	-	++	+++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
t fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	+	+	-	-	-	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	+	+	-	-	-	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	+	+	-	-	-	+
	b	-	+	++	+++	++	-	+	++	+++	++	-	+	++	++

*a = Treated with flower extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Fig.4 Effect of flower extract of Lantana camara on the development of fruit rot caused by A. niger in the presence of D. busckii.

LANTANA FLOWER

TREATED

Treated Prior to inoculation

Treated after inoculation

Treated - insect fed

A B C

LANTANA FLOWER

(Insect Fed)

Treated Prior to inoculation

A B C

Treated after inoculation

A B C

insect alone; 10 days both in inoculated with fungus prior and after treatment; 10 days when insect fed and inoculated after treatment; 7-10 days fed with insects and inoculated prior to treatment; while in 0.1% values were more or less identical in all the treatments of inoculation and insect feeding except in fungal inoculated after treatment where it was 5-10 days under various condition. It therefore, appears that with increase in dilutions the time required for (++) type of rotting decreased in most of the treatment i.e., ± 50% fruit surface was found covered in less time.

Leaf extract

Results presented in table,21 show that the fruits of tomato remained free of infection upto 15 days when treated with different concentrations of leaf extract of L. camara and allowed to feed with insect alone and uninoculated control. In fungal inoculation the incidence of rotting was almost the same when inoculation was done prior and after treatment. However, when insects were allowed to feed with fungal inoculation the rotting was more when fruits were treated with extract after inoculation and feeding. The initiation of rotting was delayed in treatment of fruits with leaf extract prior to inoculation and feeding with insect. The rotting increased with the increase in the duration of feeding. With the increase in dilution of the leaf extract, the efficacy of the extract decreased.

1. Effect of treatment of fruits with different concentrations of the leaf extract of Lantana camara on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

eatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
lated and unfed	a*	-**	-	-	-	-	-	-	-	-	+	-	-	-	-
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
ted with fungus treatment	a	+	+	+	++	++	+	+	+	++	++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ted with fungus treatment	a	-	+	+	++	++	-	-	+	++	++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ted with fungus sect fed (mts) treatment															
10	a	-	-	-	+	++	-	-	++	+++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	+	++	-	-	++	++	++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	+	+	+	++	+++	-	+	++	++	++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ted with fungus sect fed (mts) treatment															
10	a	-	-	-	+	++	-	-	+	++	+++	-	-	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	+	++	+++	-	-	+	++	++	-	-	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	-	-	+++	-	-	-	-	++	-	-	-	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

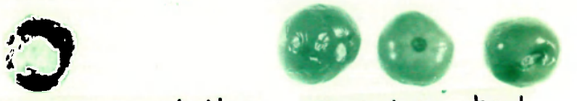
**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Fig.5. Effect of leaf extract of Lantana camara on the development of fruit rot caused by A. niger in the presence of D.busckii.

LANTANA

Treated Treated Prior to inoculation



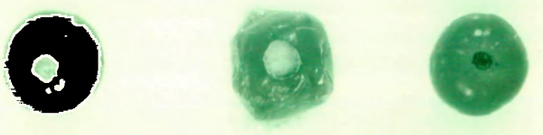
Treated after inoculation Treated - insect fed

A B C

(Insect Fed)

Treated Prior to inoculation

A B C



Treated after inoculation

A B C

The time required for development of (++) type of rotting was more than 15 days in untreated and insect fed alone; 10 days in inoculated with fungus after treatment; 7-10 days in inoculated with fungus prior to treatment; 7-15 days fungal inoculation in the presence of insect both after and prior to treatment. In this way treatment of fruits with the extract prior to inoculation was found to be more effective than after inoculation, as at 1.0% concentration 10 days were required for the development of (++) type of reaction where fruits were treated after inoculation and more than 15 days prior to inoculation in the presence of insects.

By and large the leaf extract was found more effective than the flower extract in minimising the fruit rot both in the presence or absence of insect.

4.10.4.6 Callistemon lanceolatus

Results presented in table, 22 show that there was no rotting in healthy fruits treated with standard extract of leaf of C. lanceolatus for 5 days while in those fed with insect alone for 10 and 20 minutes there was no rotting for 15 days. When fungal inoculated fruits were fed with insect for 10, 20 and 30 minutes, the rotting was comparatively less when fungal inoculation was done after treatment.

The increase in dilution decreased the efficacy. Time required for (++) type of reaction on fruits in various concentrations was 10 days in uninoculated and unfed; 7 days with

22. Effect of treatment of fruits with different concentrations of the leaf extract of Callistemon lanceolatus on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busokii

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
oculated and unfed	a*	-**	-	+	++	++	-	-	-	++	++	-	-	-	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
lated with fungus	a	-	+	++	++	++	-	+	++	+++	+++	-	+	++	+++
treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus	a	-	-	++	++	++	-	-	++	+++	+++	-	-	++	++
re treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus insect fed (mts) treatment															
10	a	-	-	++	+++	+++	-	-	++	++	++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	++	+++	+++	-	++	++	++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	++	++	++	-	++	++	++	++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus insect fed (mts) re treatment															
10	a	-	-	++	++	+++	-	++	++	++	+++	-	+++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	+	++	+++	+++	-	++	++	++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	+	++	+++	+++	-	++	++	++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ct fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	++
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	+	+	-	-	-	-
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	+	+	-	-	-	-	-	-	-	-	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

fungus inoculated (both prior and after treatment); 7 days when fed with insects for 10 minutes, 5-7 days for 20 minutes and 30 minutes after treatment along with fungal inoculation. However, when inoculation was done before treatment the time required was 5-7 days for various duration of feeding. When fruits were fed with insect alone, the time required was either 10 days or more in various feeding duration of insect, treated with different concentration of leaf extract.

4.10.4.7 Eucalyptus globulus

Rotting of tomatoes developed on 7^{th} day in 'S' concentration of the extract in all the treatments except in insect fed alone, where the rotting developed on 10^{th} day with feeding for 10 and 20 minutes. While with insect feeding for 30 minutes no rotting took place upto 15 days.

The efficacy of the extract however, decreased with the increase in dilutions. Time required for (++) type of rotting ranged from 7 days in uninoculated unfed; 5-10 days with fungus after treatment; 5-7 days with fungus before treatment; 7-15 with fungus in the presence of insect after treatment; 5-10 days with fungus before treatment and 10-15 days in insect alone (table,23).

4.10.4.8 Mentha arvensis

It is clear from table,24 that in tomato fruits treated with standard extract of leaf of M. arvensis the

23. Effect of treatment of fruits with different concentrations of the leaf extract of *Eucalyptus globulus* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila busckii*

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
culated and unfed	a*	-**	-	+	++	+++	-	-	++	++	+++	-	-	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
lated with fungus	a	-	-	+	++	++	-	++	++	+++	+++	-	++	+++	+++
treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus	a	-	-	++	++	+++	-	+	++	+++	+++	-	++	++	+++
e treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) treatment															
10	a	-	-	+	+	++	-	+	++	+++	+++	-	+	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	++	+++	-	+	++	+++	+++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	+	++	+++	-	-	+	++	++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) e treatment															
10	a	-	-	++	+++	+++	-	-	++	++	++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	+	++	+++	-	+	++	++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	+	++	++	-	+	++	+++	+++	-	++	++	+++
	b	-	+	+++	+++	++	-	+	+++	+++	+++	-	+	+++	+++
t fed alone (mts)															
10	a	-	-	-	+	++	-	-	-	+	++	-	-	-	-
	b	-	+	+	+	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	+	++	-	-	-	+	++	-	-	-	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	++
	b	-	+	++	+++	++	-	+	+	+++	++	-	+	++	++

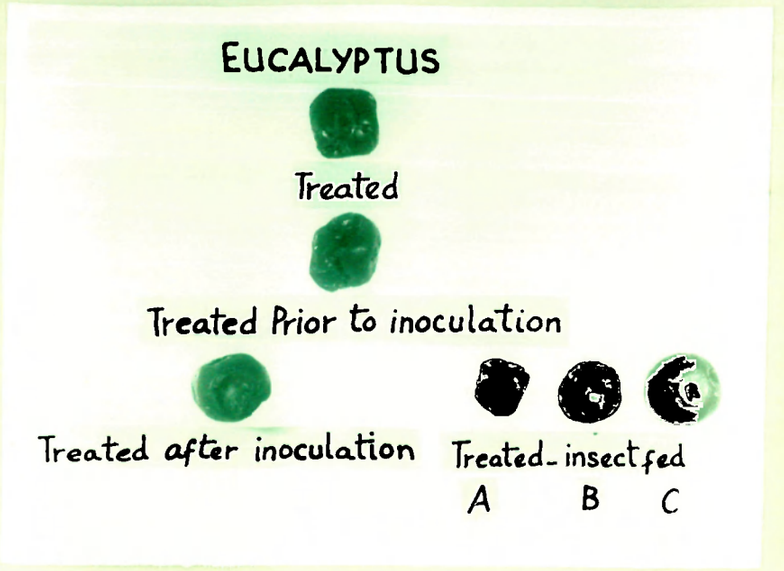
*a = Treated with leaf extract

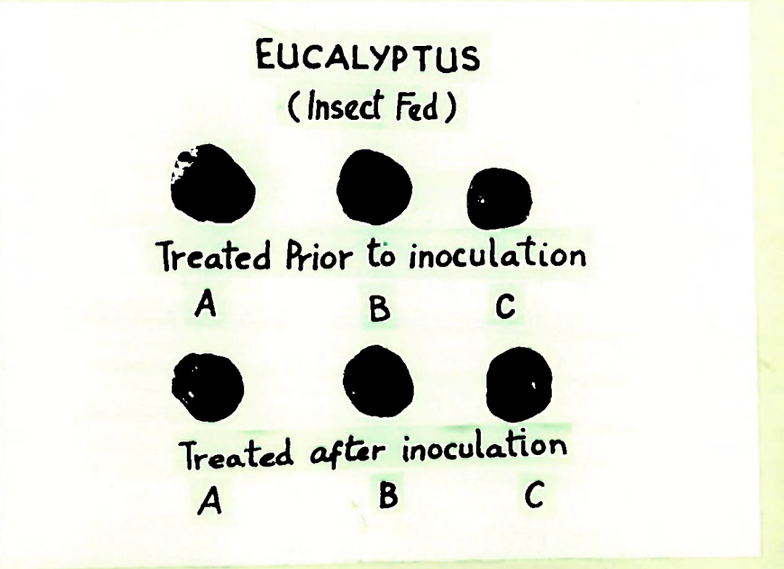
b = Control (untreated)

** (-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Fig.6 Effect of leaf extract of *Eucalyptus globulus* on the development of fruit rot caused by *A. niger* in the presence of *D. busckii*.


EUCALYPTUS
Treated
Treated Prior to inoculation
Treated after inoculation
Treated - insect fed
A
B
C


EUCALYPTUS
(Insect Fed)
Treated Prior to inoculation
A
B
C
Treated after inoculation
A
B
C

rotting was initiated after 2 days in uninoculated and unfed with insect and 10 days in those fed with insect alone. The treatment of fruits with extract before fungus inoculation was more effective than after inoculation. Similarly, the rotting was less when fruits were treated with leaf extract prior to fungal inoculation with insect feeding than with identical treatment with fruits treated after fungal inoculation.

The protection period was 5-7 days in 1% and 2-5 days in 0.1% in fungal inoculated fruits. However, in uninoculated and unfed; and insect fed alone the degree of rotting was moderate (++) in all the concentrations of the extract and poor to moderate after 10 days in insect fed alone.

Time required for 50 percent surface rotting was 7-10 days, in 'S', 1% and 0.1% in different treatments.

4.10.4.9 Ocimum sanctum

Results tabulated in table,25 show that 'standard' extract of leaf of Ocimum sanctum kept the tomato fruits free from infection for 15 days in uninoculated with fungus and unfed with insect and those fed with insect alone. When treatment with leaf extract was given both prior and after fungal inoculation the rotting was initiated after 10 days but the degree of rotting was less in pre-inoculation treatments.

Table 24. Effect of treatment of fruits with different concentrations of the leaf extract of Mentha arvensis on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
culated and unfed	a*	-**	+	++	++	++	-	++	++	++	++	-	++	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
lated with fungus treatment	a	-	-	-	-	+	-	-	+	++	+++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus e treatment	a	-	+	+	++	++	-	-	+	++	+++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) treatment															
10	a	-	-	-	+	+	-	-	-	+	++	-	-	-	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	++	++	-	-	++	++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	+	++	++	-	-	++	++	+++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
lated with fungus nsect fed (mts) e treatment															
10	a	-	-	-	+	+	-	-	-	++	++	-	-	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	+	++	+++	-	-	-	++	++	-	-	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	+	++	+++	-	-	-	-	-	-	-	-	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
t fed alone (mts)															
10	a	-	-	-	-	++	-	-	-	+	++	-	-	-	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	+	+	-	-	-	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	+	+	-	-	-	-	-	-	-	-	+
	b	-	+	++	+++	++++	-	+	++	+++	++++	-	+	++	+++

*a = Treated with leaf extract

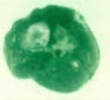
b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Fig.7 Effect of leaf extract of Mentha arvensis on the development of fruit rot caused by A. niger in the presence of D. busckii.

MENTHA



Treated

Treated Prior to inoculation

Treated after inoculation

MENTHA

(Insect Fed)

Treated Prior to inoculation

A B C

Treated after inoculation

A B C

In insect feeding for 10, 20 and 30 minutes, the rotting was also less when inoculation with fungus was done after treatment with the leaf extract.

With increase in dilution the efficacy decreased. The fruits remained free from rotting for 5-7 days in 1%, 2-5 days in 0.1% in inoculated fruits. Time required for (++) type of fruit rot was >15 days in uninoculated and unfed and 15 days in fungal inoculated after treatment with leaf extract but it was reduced to 10 days when inoculated with fungus before treatment in 'S' conc. In insect feeding with fungal inoculation it ranged from 7-15 days in different concentration and feeding durations. In insect fed alone it was either 10 or more than 15 days.

Therefore, with increase in dilution of extract the time required for (++) type of rotting decreased in most of the treatment.

4.10.4.10 Adenocalymna alliacea

As a result of treatment with leaf extract there has been no rotting of fruits for 7 days in uninoculated with fungus and unfed with insect in 'S', 1% and 0.1% and upto 15 days when fed with insect only for 10 minutes and 5-7 days when fed with insect for 20 and 30 minutes with 'S' concentration. Rotting of fruits when inoculation was done either before or after treatment with 'S' took place after 7 days and intensity

25. Effect of treatment of fruits with different concentrations of the leaf extract of *Ocimum sanctum* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila buscki*

eatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
ulated and unfed	a*	-**	-	-	-	-	-	-	+	++	+++	-	-	-	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
ated with fungus treatment	a	-	-	-	+	++	+	++	++	++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
ated with fungus treatment	a	-	-	-	++	+++	-	-	-	++	++	-	+	+	++
	b	-	+	++	++	+++	-	+	++	++	+++	-	+	++	+++
ated with fungus sect fed (mts) treatment															
10	a	-	-	-	++	+++	-	-	++	++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	-	++	+++	-	-	++	++	++	-	-	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	+	+	+	+++	-	+	++	+++	+++	-	-	++	++
	b	-	+	++	+++	++	-	+	++	+++	+++	-	+	++	+++
ated with fungus sect fed (mts) treatment															
10	a	+	+	+	++	+++	-	-	-	+	++	-	+	++	++
	b	-	+	+++	+++	+++	-	+	++	++	+++	-	+	+++	++
20	a	-	+	+	++	+++	-	+	+	++	++	-	+	++	++
	b	-	+	+++	+++	+++	-	+	+++	++	+++	-	+	+++	++
30	a	-	+	+	++	+++	-	+	+	++	++	-	+	++	++
	b	-	+	++	+++	+++	-	+	+++	++	+++	-	+	+++	++
fed alone (mts)															
10	a	-	-	-	-	+	-	-	-	-	-	-	-	-	++
	b	-	+	+	+	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	-	-	-	-	-	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	+	-	-	-	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Fig.8 Effect of leaf extract of Ocimum sanctum on the development of fruit rot caused by A. niger in the presence of D. busckii.

OCIMUM

Treated

Treated Prior to inoculation

Treated after inoculation

OCIMUM

(Insect Fed)

Treated Prior to inoculation

A B C

Treated after inoculation

A B C

has been almost the same. But in those inoculated with fungus in the presence of insects for different intervals and treated with extracts there has been some difference in treatments with 'S' concentration in before and after treatments. The rotting took place only after 15 days when the insects were allowed to remain for 10 minutes over the fruits inoculated with fungus after treatment, whereas in corresponding duration of insect feeding, the rotting took place after 7 days when inoculated with fungus before treatment. At other feeding intervals there has been no difference between the two kinds of treatments (i.e., treatment before/after inoculation). The efficacy of the leaf extract in minimising the rotting decreased with increase in dilution of the extract (table, 26).

4.10.4.11 Chenopodium album

Results presented in table, 27 show that there was no rotting of fruits upto 5 days in all the treatments except in insect fed alone where the rotting did not occur for 15 days and insect fed for 10 minutes with fungus inoculation. There has been no difference in rotting in 'S' concentration in inoculations before and after treatment. Similarly there has been less rotting in the presence of insects for different duration, when inoculation with fungus was done after the treatment. However, there has been no rotting for 15 days in insect fed alone as a result of treatment with leaf-extract.

e 26. Effect of treatment of fruits with different concentrations of the leaf extract of *Adenocalymna alliacea* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila busckii*

reatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	
oculated and unfed	a*	-**	-	-	++	+++	-	-	-	++	++	-	-	-	++	+
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++	+
ulated with fungus r treatment	a	-	-	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
ulated with fungus re treatment	a	-	-	++	++	+++	-	+	++	+++	+++	-	+	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
ulated with fungus insect fed (mts) r treatment																
10	a	-	-	-	-	+	-	+	++	++	+++	-	+	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
20	a	-	-	++	++	+++	-	+	++	+++	+++	-	+	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
30	a	-	-	++	++	+++	-	+	++	+++	++	-	+	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	++	-	+	++	+++	+
ulated with fungus and ct fed (mts) re treatment																
10	a	-	-	++	++	+++	-	++	++	++	+++	-	+	++	+	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	++	+
20	a	-	-	++	+++	+++	-	++	++	++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	++	+++	+
30	a	-	-	++	++	+++	-	++	++	++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
ct fed alone (mts)																
10	a	-	-	-	-	-	-	-	-	++	++	-	-	-	++	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	+
20	a	-	-	+	++	+++	-	-	-	++	++	-	-	++	++	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	+
30	a	-	-	-	++	+++	-	-	-	++	+++	-	-	++	++	+
	b	-	+	+	++	+++	-	+	++	++	+++	-	+	++	++	+

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Time required for the development of fruit rot to the extent of (++) ranged from 5-10 days; except those where the fruits remained free from insects for about 15 days, in various concentrations of the extract and in various treatments.

4.10.4.12 Peucedenum graveolans

There has been no fruit rot in healthy fruits for 15 days when treated with 'S' concentration of leaf extract for 10 days and in those fed with insect alone for 30 minutes. Rotting, however, was initiated after 5 days when inoculated with fungus after treatment; 2 days when inoculated with fungus before treatment; in insect feeding for 10 minutes after treatment the fruits remained free from infection upto 10 days. Increase in duration brought an early initiation of the disease; no fruit rot for 5 days when feeding was for 20 and 30 minutes. In corresponding insect feeding with treatment after fungal inoculation there was no rot for 2 days in all the feeding durations. In insect alone, however, there was no rotting for 7 days in 10 and 20 minutes feeding. Therefore, treatment before inoculation was more effective than after.

Increasing dilutions resulted in decrease in its efficacy. Time required for 50% fruit rot (++) was 5-7 days in all the treatments except in insect fed alone where it ranged from 10-15 days in different concentrations and feeding durations (table,28).

le 27. Effect of treatment of fruits with different concentrations of the leaf extract of Chenopodium album on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buskii

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
noculated and unfed	a*	-**	-	+	++	+++	-	-	+	++	++	-	-	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
culated with fungus	a	-	-	++	++	+++	-	+	++	++	+++	-	+	++	+++
er treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus	a	-	-	++	++	++	-	+	++	++	++	-	+	++	++
ore treatment	b	-	+	++	+++	++	-	+	++	+++	++	-	+	++	+++
culated with fungus insect fed (mts) er treatment															
10	a	-	-	-	-	-	-	-	++	++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	++	++	++	-	-	++	+++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	++	+++	+++	-	-	++	+++	+++	-	-	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
culated with fungus insect fed (mts) ore treatment															
10	a	-	-	-	++	++	-	++	++	+++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	++	++	+++	-	++	++	+++	+++	-	++	+++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	++	++	+++	-	-	++	+++	+++	-	++	+++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ect fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	-	-	-	+	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	-	-	-	-	+	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	+	+	++	-	-	+	+
	b	-	+	++	+++	++	-	+	++	+++	++	-	+	++	++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

able 28. Effect of treatment of fruits with different concentrations of the leaf extract of *Peucedenum graveolans* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila buscki*

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
ninoculated and unfed	a*	-**	-	-	-	+	-	+	++	++	+++	-	-	++	++
	b	-	-	+	+++	++	-	-	+	+++	+++	-	-	+	++
noculated with fungus fter treatment	a	-	-	++	++	+++	-	++	+++	+++	++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	++	-	+	++	++
noculated with fungus efore treatment	a	-	++	++	++	+++	-	++	++	+++	++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++
noculated with fungus nd insect fed (mts) fter treatment															
10	a	-	-	-	-	+	-	+	++	+++	+++	-	++	+++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	++	+++	-	+	++	+++	+++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	++	++	++	-	+	++	+++	+++	-	++	++	+++
	b	-	+	++	+++	++	-	+	++	+++	+++	-	+	++	+++
noculated with fungus nd insect fed (mts) efore treatment															
10	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	++	+++
20	a	-	+	+++	+++	+++	-	++	++	+++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	++	+++
30	a	-	+	+++	+++	+++	-	++	++	+++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	++	+++
nsect fed alone (mts)															
10	a	-	-	-	++	+++	-	-	+	++	+++	-	-	+	++
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	++	+++	-	-	-	++	+++	-	-	+	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	++	-	-	+	++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

4.10.4.13 Foeniculum vulgare

Leaf extract

Results presented in table, 29 show that no rotting of fruits was observed for 7 days in those uninoculated with fungus and unfed with insect and upto 15 days with insect fed alone. When treated with 'S' concentration of the extract of F. vulgare there has been no difference in the rotting of fruits in treatment with leaf extract both prior and after fungal inoculation. In both, the rotting was seen only after 7 days. When insects were allowed to feed for 10, 20 and 30 minutes, the rotting was less when fungal inoculation was done after the treatment with extract. The efficacy of the leaf extract in minimising the rotting decreased with the increase in dilution of the extract.

Time required for the development of rotting to the extent of ± 50 percent fruit cover (++) was 7-10 days in 'S'; 5-7 days in 1% and 0.1%.

Flower extract

Almost identical results were obtained when fruits were treated with flower extract except that when insects were allowed to feed for 10 minutes after fungal inoculation (in fruits treated prior to fungal inoculation), the initiation of rotting was delayed in flower extract treated ones and also when fed with insects for different durations after fungus

Table 29. Effect of treatment of fruits with different concentrations of leaf extract of *Foeniculum vulgare* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila buscki*

Treatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
Uninoculated and unfed	a*	-**	-	-	++	+++	-	-	++	++	+++	-	-	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++
Inoculated with fungus after treatment	a	-	-	++	++	+++	-	+	++	+++	+++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
Inoculated with fungus before treatment	a	-	-	++	++	+++	-	-	++	+++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
Inoculated with fungus and insect fed (mts) after treatment															
10	a	-	-	+	++	+++	-	+	++	+++	+++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
20	a	-	-	+	++	+++	-	+	++	+++	+++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
30	a	-	-	+	++	++	-	+	++	+++	+++	-	+	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++
Inoculated with fungus and insect fed (mts) before treatment															
10	a	-	-	++	+++	+++	-	++	++	+++	+++	-	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	++	+++	+++	-	++	++	+++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	++	++	+++	-	++	++	+++	+++	-	++	+++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
Insect fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	+	+	-	-	++	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

inoculation (in fruits treated after fungus inoculation) the onset of rotting was on 5th day in flower extract treated fruits as against 7 days in leaf extract treated fruits (table, 30).

4.10.4.14 Allium cepa and Allium sativum

Allium cepa and A. sativum were tested for their efficacy for controlling fruit rot caused by A. niger both in the presence and absence of insect (tables 31,32). In uninoculated and unfed with insect the fruit rot developed after 7 days when treated with A. cepa extract while after 10 days with A. sativum. In those inoculated with fungus alone after treatment with leaf extract rotting was observed after 7 days in both but when inoculated before treatment the rotting occurred after 7 days with A. sativum and 10 days with A. cepa. In the presence of insect when inoculation with fungus was done the rotting was observed on 7th day when treated with A. cepa and 5th day with A. sativum. Thus, A. cepa, appeared more effective for insects both in those treated before and after inoculation. But the intensity of rotting was more in those treated with A. sativum. Similarly, in those fed with insect alone there has been no rotting upto 10 days when treated with A. sativum and >15 days with A. cepa.

It, therefore, appears that extract of A. cepa is more effective than A. sativum in minimising the fruit rot of tomato.

e 31. Effect of treatment of fruits with different concentrations of the leaf extract of Allium cepa on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

reatments		Rotting of fruits in different concentration after (days)													
		2	5	7	10	15	2	5	7	10	15	2	5	7	10
oculated and unfed	a*	-**	-	++	++	+++	-	+	++	++	+++	-	-	++	++
	b	-	-	+	+++	+++	-	-	+	++	++	-	-	+	+++
ulated with fungus r treatment	a	-	-	+	++	+++	-	+	++	++	+++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	+++
ulated with fungus re treatment	a	-	-	-	++	++	-	+	++	++	++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	+++
ulated with fungus insect fed (mts) r treatment															
10	a	-	-	++	++	+++	-	+	++	++	+++	-	++	++	++
	b	-	+	++	++	+++	-	+	++	++	+++	-	+	++	++
20	a	-	-	++	++	+++	-	++	++	++	+++	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	++
30	a	-	-	++	+++	+++	-	++	++	++	+++	-	++	++	+++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	+++
ulated with fungus insect fed (mts) re treatment															
10	a	-	-	++	++	+++	-	++	++	++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
20	a	-	-	++	+++	+++	-	++	++	++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
30	a	-	-	++	++	+++	-	++	++	++	+++	-	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++
ct fed alone (mts)															
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++
20	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	++	-	+	++	+++	++	-	+	++	+++

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

e 30. Effect of treatment of fruits with different concentrations of flower extract of Foeniculum vulgare on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

reatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	1
oculated and unfed	a*	-**	-	-	++	+++	-	-	-	++	++	-	-	-	++	+
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++	+
lated with fungus	a	-	+	++	++	+++	-	-	++	+++	+++	-	-	++	++	+
treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++	+
lated with fungus	a	-	-	++	++	+++	-	-	++	+++	+++	-	++	++	++	+
re treatment	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++	+
lated with fungus																
nsect fed (mts)																
treatment																
10	a	-	-	-	-	-	-	-	++	+++	+++	-	-	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
20	a	-	-	+	++	+++	-	-	++	+++	+++	-	-	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
30	a	-	+	++	+++	+++	-	++	++	+++	+++	-	++	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
lated with fungus																
nsect fed (mts)																
e treatment																
10	a	-	+	++	+++	+++	-	++	+++	+++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	++	+
20	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	+++	+++	+
30	a	-	+	++	++	++	-	++	++	+++	+++	-	++	++	++	+
	b	-	+	+++	+++	++	-	+	+++	+++	+++	-	+	+++	++	+
t fed alone (mts)																
10	a	-	-	-	+	++	-	-	+	++	+++	-	-	++	++	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	+
20	a	-	-	-	-	-	-	-	+	++	++	-	-	++	++	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	+
30	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	++	+++	++	-	+	++	++	+++	-	+	++	+++	+

*a = Treated with flower extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

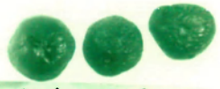
Fig.9 Effect of leaf extract of *Allium cepa* on the development of fruit rot caused by *A. niger* in the presence of *D. busckii*.

ONION

TREATED

Treated Prior to inoculation

Treated after inoculation



Treated - insect fed

A B C

ONION

(Insect Fed)

Treated Prior to inoculation

A B C

Treated after inoculation

A B C

e 32. Effect of treatment of fruits with different concentrations of the leaf extract of *Allium sativum* on the development of fruit rot of tomato caused by *Aspergillus niger* in the presence of *Drosophila buscckii*

Treatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	1
oculated and unfed	a*	-**	-	-	++	+++	-	-	-	++	++	-	-	-	++	+
	b	-	-	+	+++	++	-	-	+	+++	+++	-	-	+	++	+
ulated with fungus r treatment	a	-	-	+	++	+++	-	-	+	++	+++	-	++	++	++	+
	b	-	+	++	+++	++	-	+	++	+++	+++	-	+	++	++	+
ulated with fungus re treatment	a	-	-	++	+++	+++	-	-	++	++	++	-	++	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++	+
ulated with fungus insect fed (mts) r treatment																
10	a	-	+	+	++	+++	-	-	+	++	+++	-	-	++	+++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
20	a	-	+	+	++	+++	-	+	++	++	++	-	+	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
30	a	-	+	+	++	+++	-	+	++	++	+++	-	+	++	+++	+
	b	-	+	++	+++	++	-	+	++	+++	+++	-	+	++	+++	+
ulated with fungus insect fed (mts) re treatment																
10	a	-	-	++	++	+++	-	++	++	++	+++	-	++	++	++	+
	b	-	+	+++	+++	++	-	+	+++	+++	+++	-	+	+++	+++	+
20	a	-	++	++	+++	+++	-	++	++	++	+++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
30	a	-	++	++	+++	+++	-	++	++	++	++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
ct fed alone (mts)																
10	a	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	+
20	a	-	-	-	-	+	-	-	-	-	++	-	-	-	-	-
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	+
30	a	-	-	-	-	+	-	-	-	-	++	-	-	-	-	-
	b	-	+	++	+++	++	-	+	++	+++	++	-	+	++	+++	+

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

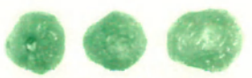
Fig.10 Effect of leaf extract of *Allium sativum* on the development of fruit rot caused by *A. niger* in the presence of *D. busckii*.

GARLIC

TREATED

Treated Prior to inoculation

Treated after inoculation



Treated - insect fed

A B C

GARLIC

(Insect Fed)

Treated Prior to inoculation

A B C

Treated after inoculation

A B C

In both, the leaf extract dilution reduced the efficacy.

Time required for (++) type of reaction was 5-10 days in fungus inoculated after treatment; 5-7 days before treatment, 7-10 days in the presence of insect for 10 minutes, 20 and 30 minutes,, after treatment, 5-7 days with 10 minutes and 5 days with 20 and 30 minutes before treatment in different concentrations of the leaf extract of A. sativum. The values with A. cepa were 7 days in uninoculated and unfed; 5-10 days with inoculated before and after treatment; 5-7 days with insect for different duration before and after treatment and in remaining 15 days or more for different conc. of extract.

4.10.4.15 Cymbopogon citratum

In treatment with leaf extract there has been no rotting for 10 days in fruits treated but uninoculated and unfed; and fungal inoculated both prior and after treatment. When fed with insect alone for 10 minutes, no rotting occurred upto 15 days. There has been thus no difference in the rotting of fruits when treatment was done either prior or after fungal inoculation. Similarly, the treatment of fruits with extract either prior or after fungal inoculation in the presence of insect made no difference in the extent of rotting The degree of protection provided by the extract, however, decreased with the increase in dilution of the extract.

Time required for the development of 50% (++) type of rotting was 10-15 days in unfed and uninoculated, 7-15 days in fungal inoculated before treatment; 10 days in fungal inoculated after treatment; 7-15 days in insect fed various duration and fungal inoculated after treatment, 5-10 days in insect fed for various duration and fungal inoculated before treatment and 7 to >15 days in insect fed alone in three concentrations of the leaf extract, It is, therefore, clear that in the presence of insect in fungal inoculation after treatment the rate of disease development is low (table,33).

4.10.5 Effect of latex of Euphorbia hirta and Calotropis procera on development of fruit rot caused by A. niger in presence of D. busckii

4.10.5.1 Euphorbia hirta

When fruits were treated with 'S' conc. of latex of Euphorbia hirta, the fruit rot development was observed after 10 days in uninoculated and unfed; 7 days in inoculated with fungus after treatment; 5 days in inoculated with fungus before treatment; 7 days with fungus inoculation in the presence of insect for different duration after treatment; 5 days with fungus inoculation in the presence of insect before treatment and 10-15 days in insect fed alone. In all the above cases, the fruit rot onset was delayed as a result of treatment with latex. With the increase in dilution the efficacy of the latex decreased.

e 33. Effect of treatment of fruits with different concentrations of the leaf extract of Cymbopogon citratum on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buscki

reatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	1
oculated and unfed	a*	-**	-	-	-	++	-	-	-	+	++	-	-	-	++	+
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++	+
ulated with fungus r treatment	a	-	-	-	-	++	-	-	+	++	+++	-	-	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
ulated with fungus re treatment	a	-	-	-	-	++	-	-	+	+	++	-	-	-	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	++++	+
ulated with fungus insect fed (mts) r treatment																
10	a	-	-	++	+++	+++	-	-	++	++	++	-	-	++	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
20	a	-	-	+	++	+++	-	-	-	++	++	-	-	-	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
30	a	-	-	+	++	+++	-	-	-	++	++	-	-	-	++	+
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+
ulated with fungus insect fed (mts) re treatment																
10	a	-	-	++	+++	+++	-	+	++	++	++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
20	a	-	+	+	++	+++	-	-	+	++	+++	-	+	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
30	a	-	+	++	++	+++	-	-	++	++	++	-	++	++	++	+
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+
ct fed alone (mts)																
10	a	-	-	-	-	-	-	-	-	-	+	-	-	-	-	+
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	+
20	a	-	-	-	+	+	-	-	+	+	+	-	-	+	+	+
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	+
30	a	-	-	-	-	++	-	-	+	++	++	-	-	++	++	+
	b	-	+	++	+++	++++	-	+	++	++++	++++	-	+	++	++++	+

*a = Treated with leaf extract

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

For (++) type of reaction to develop less time was required in latex treated fruits. In uninoculated and unfed; inoculated with fungus after treatment both in the presence and absence of insect it was 10 days in treated as against 7 days in untreated. In presence of insect before treatment it was 7 days as against 5 days in untreated, and in insect fed alone; and 15 days as against 7-10 days in untreated (table, 34).

4.10.5.2 Calotropis procera

Results presented in table, 35 show that fruits treated with latex of Calotropis procera remained free upto 15 days in uninoculated and unfed with insect and free from rot upto 15 days when fed with insect for 10 minutes; but the rotting ranged from poor to severe when insect feeding period was prolonged to 20-30 minutes. There has been practically no difference in the rotting of fruits when treatment was given prior or after fungal inoculation but it was less than untreated controls.

With increase in dilution not only the protection period decreased but intensity of rotting was also increased. Time required for the development of rotting to the extent of (++) type of reaction was 5-7 days in fungal inoculations, 7-10 days in fungal inoculation with insect feeding after treatment; 5-7 days in fungal inoculation with insect feeding before treatment; and 7-15 days with insect alone in different concentrations of the latex.

Table 34. Effect of treatment of fruits with different concentrations of the latex of Euphorbia hirta on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila buskii

Treatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	15
Uninoculated and unfed	a*	-**	-	-	++	+++	-	++	++	++	++	-	++	++	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++	+++
Inoculated with fungus after treatment	a	-	-	+	+++	+++	-	++	++	++	+++	-	++	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus before treatment	a	-	+	++	++	+++	-	++	++	++	++	-	++	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus and insect fed (mts) after treatment																
10	a	-	-	+	++	+++	-	+	++	++	++	-	+	++	++	++
	b	-	+	++	+++	+++	-	+	++	++	+++	-	+	++	+++	+++
20	a	-	-	+	++	++	-	+	++	+++	+++	-	+	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
30	a	-	-	+	++	+++	-	+	++	++	++	-	+	++	+++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus and insect fed (mts) before treatment																
10	a	-	+	++	++	+++	-	++	++	++	++	-	++	+++	+++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+++
20	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+++
30	a	-	-	-	-	-	-	++	+++	+++	+++	-	++	++	++	+++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	+++
Insect fed alone (mts)																
10	a	-	-	-	+	++	-	-	-	++	++	-	-	-	++	++
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	++
20	a	-	-	-	+	++	-	-	-	++	++	-	-	-	++	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	++
30	a	-	-	-	-	-	-	-	-	++	++	-	+	++	++	+++
	b	-	+	++	+++	+++	-	+	++	++	++	-	+	++	++	+++

*a = Treated with latex

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

Table 55. Effeot of treatment of fruits with different concentrations of the latex of Calotropis prooera on the development of fruit rot of tomato caused by Aspergillus niger in the presence of Drosophila busckii

Treatments		Rotting of fruits in different concentration after (days)														
		2	5	7	10	15	2	5	7	10	15	2	5	7	10	15
Uninoculated and unfed	a*	-**	-	-	-	-	-	-	++	++	++	-	+	++	++	++
	b	-	-	+	+++	+++	-	-	+	+++	+++	-	-	+	+++	+++
Inoculated with fungus after treatment	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	+++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus before treatment	a	-	+	++	++	+++	-	++	++	++	+++	-	++	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus and insect fed (mts) after treatment																
10	a	-	+	+	++	+++	-	+	++	++	+++	-	+	++	++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
20	a	-	+	+	++	+++	-	+	+	++	++	-	+	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
30	a	-	+	+	++	+++	-	+	++	+++	+++	-	+	++	+++	+++
	b	-	+	++	+++	+++	-	+	++	+++	+++	-	+	++	+++	+++
Inoculated with fungus and insect fed (mts) before treatment																
10	a	-	+	++	++	+++	-	-	++	++	++	-	++	++	+++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	++
20	a	-	+	++	++	+++	-	++	++	++	+++	-	++	+++	+++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	++
30	a	-	+	++	++	+++	-	++	++	+++	+++	-	++	++	++	++
	b	-	+	+++	+++	+++	-	+	+++	+++	+++	-	+	+++	+++	++
Insect fed alone (mts)																
10	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	b	-	+	+	++	++	-	+	+	++	++	-	+	+	++	++
20	a	-	+	+	+	++	-	-	+	+	++	-	-	-	++	++
	b	-	+	+	+	++	-	+	+	+	++	-	+	+	+	++
30	a	-	-	+	++	+++	-	-	++	++	++	-	-	++	++	++
	b	-	+	++	+++	+++	-	+	++	+++	++	-	+	++	++	++

*a = Treated with latex

b = Control (untreated)

**(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates.

4.10.6 Effect of growing tomato seedlings in soil treated with different oil cakes and bavistine on the development of fruit rot when inoculated with Aspergillus niger in the presence of Drosophila busckii .

The fruits of tomato obtained from plants grown in soil amended with oil cakes and bavistine remained relatively free of rotting for about 15 days when inoculated with A. niger both in presence and absence of insect or insect alone except in those obtained from plants grown in soil amended with castor cake + bavistine and cakes of castor, mustard + bavistine, where rotting occurred after 7 days of inoculation of fungus. In fruits obtained from plants grown in unamended soil the rotting occurred on 5^{th}- 7^{th} day when inoculated with fungus in presence or absence of insect or insect fed alone (table,36).

Table 36. Effect of growing tomato seedlings in soil treated with oil cakes and bavistine on the development of fruit rot when inoculated with Aspergillus niger in presence of Drosophila busckii

Treatment of soil	Rotting of fruits after (days)														
	Insect					Insect + Fungus					Fungus				
	2	5	7	10	15	2	5	7	10	15	2	5	7	10	15
Castor cake	-	-	-	-	-	-	-	-	-	+	-	-	-	-	++
Mustard cake	-	-	-	-	+	-	-	-	-	+	-	-	-	-	+
Castor cake + mustard cake	-	-	-	-	++	-	-	-	+	++	-	-	-	+	++
Castor cake + bavistine	-	-	-	-	+	-	-	-	-	+	-	-	++	++	+++
Mustard cake + bavistine	-	-	-	-	+	-	-	-	-	+	-	-	-	-	++
Cakes of castor and mustard + bavistine	-	-	-	-	+	-	-	-	-	+	-	-	++	++	++
Bavistine	-	-	-	-	-	-	-	-	-	+	-	-	-	+	+
Control (untreated)	-	-	++	++	+++	-	-	++	++	+++	-	-	++	++	+++

(-) Nil, (+) poor, (++) moderate and (+++) severe

Each value is mean of 3 replicates

CHAPTER V

DISCUSSION

Fruit rot of tomato due to Aspergillus niger is very common in shops and warehouses in Aligarh. These shops and warehouses are also visited by several insects, most common of which is Drosophila busckii. In the shops and warehouses where the rotting was very severe, there was frequent visits of the insect. It was not clear if there is any relationship between the two organisms in the severity of rotting. The present studies are, therefore, aimed to work out the relationship between Aspergillus niger and Drosophila busckii in the severity of the fruit rotting caused by the former.

During survey for insects visiting the shops in various market at Aligarh, the frequency of occurrence of D. busckii has been high in amongst different insects. Maxwell (1984) has also pointed out this insect is a frequent visitor in large number on ripened fruits. In the present studies the frequency of the insect has been more during rains which is again in conformity with Maxwell (1984). Aspergillus niger has also been frequently found associated with fruit rot in amongst various fungi isolated from rotted fruits. A. niger has earlier been reported as causing fruit rot of tomato during storage (Singh and Kainsa, 1983). In order to

ascertain whether the isolate of the fungus obtained locally from the market has been pathogenic or not, inoculation test has been made in the laboratory. This isolate was found pathogenic. Therefore, in subsequent studies the isolate has been used.

The presence of inoculum of the fungus has been detected on all the parts of the insect body, indicating, thereby, that inoculum is being carried through all the parts of the body during their hopping from one fruit to other or one place to other. These results are thus in accordance with those obtained by Evans,(1973), Coyle, (1975), Dakwa, (1977), Hasan, (1982), Swincer,(1984), Verma and Pathak,(1984), Moser,(1985), Haung and Harper,(1985) and Haung et al.,(1986). The possibility of the insect providing wounds on fruits during transmission of fungal inoculum,however, can not be ruled out as reported earlier by Christensen,(1953).

During the survey of fungi associated with tomato fruits in the market at Aligarh, in all thirteen fungi have been isolated from fruits; where all the fungi have been found to exhibit higher frequency values except Actinomucor sp., Alternaria tenuis, Alternaria solani, Cladosporium fulvum, Curvularia sp, Geotrichum candidum, Fusarium sp.

Of all these fungi the frequency of A. niger has been highest in all the localities surveyed throughout the year, It is not surprising because A. niger has been found associated

with large number of storage products (Prakash et al., 1974; Prasad and Bilgrami, 1973; Singh, 1974; Singh and Kainsa, 1983; Adisa, 1985; Mandal and Das Gupta, 1984). Therefore, in the subsequent study A. niger has been used.

In order to determine whether the fungi isolated from the fruits collected from the market are the same from the fruits of standing crop or they are contracted during transport and storage, the fruits were bagged with butter paper right after the fruit setting. The results show that most of the fungi isolated are saprophytes aquired during transport and storage and not parasites, aquired during plant growth.

Studies dealing with the effect of temperature on the development of fruit rot of tomato in the presence of the insect, D. busckii show that highest rotting has been observed at 30°C, but in the presence of the insect its onset has been earlier. This temperature has been reported as optimum for growth of A. niger (Cochrane, 1958 and Denerall, 1965). These studies are in accordance with Harter and Weimer, (1922); Adams, (1923); Smoot and Segall, (1963); Tandon and Misra, (1969); Tandon and Ghosh, (1962); Prasad and Bilgrami, (1973); Mehta et al., (1975). Similarly, the rotting has been highest at 95 and 100% relative humidity, where early onset of rotting has been observed. High humidity is conducive for the rapid growth of the fungus and sporulation which might the reason for high intensity of the rotting (Tandon and Singh, 1969; Sumbali and Mehrotra, 1983; Singh et al., 1983). Presence of

D. busckii has been found to aggravate the fruit rotting to bring about early onset of rotting. Insect during bruising might be secreting saliva containing wide variety of substances and enzymes (Ross et al., 1982). These enzymes probably act on the substratum on the surface of the fruit to produce substances stimulatory to fungus. Carbohydrases and Peptides (Wigglesworth, 1979) have been reported from the saliva of certain species of Drosophila.

The ascorbic acid content decreases as a result of inoculation with fungus and feeding with insect. It is known that L-ascorbic acid is oxidised to dehydro-L-ascorbic acid by ascorbic acid oxidase or by certain other oxidative enzymes like polyphenol oxidase, cytochrome oxidase etc, (Wood, 1967). It is likely that ascorbic acid oxidase increases which metabolizes ascorbic acid in the fruit resulting in its low amount. The metabolites of the fungus and the enzymes and other substances contained in the saliva of the insect might probably be stimulating the ascorbic acid oxidase resulting in the reduction. Similar results have been obtained by Ghosh et al.,(1965) with A. niger, R. nigricans, B. theobromae, Colletotrichum papayae and Gleosporium papayae; by Ghosh et al., (1966) with Alternaria tenuis, Chaetomium globosum, Curvularia lunata, Cylindrocarpon tonkinense, Fusarium oxysporum and Helminthosporium specifera; by Tandon (1970) with Aspergillus niger; by Prasad (1977); Prasad and Prasad (1977) with Dreschlera austrialianse.

Studies on the changes in the amino acid content show that the number of amino acids in the fruits has been more in those infected with fungus and fed with insects either separately or together. This is understandable because as a result of infection, there is increase in the enzymes such as glutamine dehydrogenases and synthetases which result in production of higher amount of amino acids. Moreover, the peptidases and proteinases attack the proteins resulting in more of amino acids (Shaw and Colotelo, 1961). These results are, therefore, in agreement with those of Stretch and Capellinii,(1965) and Pandey,(1985). Absence of methionine in the insect fed and fungus inoculated could be due to susceptibility of the tomato fruit; as methionine has been suggested to be present in high concentration in the resistant plant (Wood, 1967). The possibility of enzymes released by the feeding of insect stimulating the enzymes responsible for break down of proteins, however, cannot be ruled out. The amino acids released by the fungus in the host tissues may also be contributing towards the variation in the amino acid content in the fruits infected with fungus alone (Wood, 1967). Complete absence of certain amino acids in fungus infected fruits may be attributed to their utilization by the fungus.(Bhargava and Arya, 1983) Appearance of certain amino acids in infected fruits has also been observed by Tandon (1967).

In order to reduce the degree of fruit rotting, extracts of nineteen plant species belonging to eighteen genera have been

tested against fruit rot caused by Aspergillus niger both in the presence or absence of insects. Almost extracts of all the plants have been effective in delaying the onset of fruit rot in the presence or absence of insect to a varying degree. Ethanol and ethanolic extract of L. camara, O. sanctum and M. arvensis have been found effective in controlling fruit rot as evidenced by the time required for the development of (++) type of reaction. Ethanol itself is antiseptic and a good solvent of many organic chemicals. Thus, the principles responsible for control in these plants are probably dissolved in ethanol and together with ethanol give protection to tomato fruits against fruit rot and insect. But when leaf powder, ethanol and water extract of neem, Azadirachta indica, have been compared for their efficacy, the water extract has been found more effective. It appears that the solubility of active principle(s) is more in water.

The time required for the development of (++) type of rotting in all the treatments viz., unfed-uninoculated, inoculated with fungus before and after treatment and insect fed alone (++) has been 15 days or more when treated with different preparations of neem (Azadirachta indica) except in inoculation with fungus in the presence of insect before and after treatment. When treated with extracts of Lantana camara time required for (++) type of reaction has also been ± 15 days in treatment in uninoculated and unfed, fungus inoculated in the presence of insect before and after treatment and insect fed alone; with

Cymbopogon citratum in uninoculated/unfed, inoculated with fungus before treatment, inoculated with fungus in the presence of insect after treatment and insect fed alone. Thus these plants can be rated as very effective in reducing the fruit rot caused by A. niger both in the presence or absence of insect. In fruits fed with insect alone the time required for (++) type of reaction has been ± 15 days when treated with extract of almost all the plants except Adenocalymna alliacea, Solanum xanthocarpum, Ocimum sanctum, Callistemon lanceolatus and Peucedenum graveolans. In uninoculated and unfed in many cases the fruits developed (++) type of reaction earlier than 15 days as against ± 15 days in insect fed alone probably the insects are free of inoculum or if they bear the inoculum the plant extracts provide protection against the invasion by either repelling the insect or making the inoculum ineffective. The possibility that the secretions of insects in the presence of extracts might be adversely affecting the fungus inoculum to infect the fruits, however, cannot be ruled out. This explains in part as to why the extract of plants which are effective in insect alone, have not be found very effective in uninoculated and unfed.(Tables 37,38)

Another category of plants where the application of extracts to fruits could delay the development of (++) type of reaction to 10 days include Eucalyptus globulus for uninoculated and unfed; Adenocalymna alliacea, Allium sativum, Solanum xanthocarpum, Argemone mexicana, Ocimum sanctum, Callistemon lanceolatus, Foeniculum vulgare, Cassia fistula,

tments/ of plant	'A' (5 - 7 days)	'B' (10 days)	'C' (15 days)	'D' (>15 days)
oculated and d	Al.cepa, Eup.hirta, Euc. globulus, Wi.somnifera, Cl.procera, Fo.vulgare,(L), M.arvensis, Peu.graveolans	Ad.alliacea, Al.sativum So.xanthocarpum, Ar.mexicana, O.sanctum, Cli.lanceolatus, Fo.vulgare,(F), Casi.fistula, Che.album	Cy.citratum.	L.camara,(L), Az.indica.(D) (A)
ulated with us after tment	Ad.alliacea, Al.cepa, Eup-hirta, Euc. globulus, Wi. somnifera, So.xanthocarpum, Ar.mexicana, Cl.procera, Cli.lanceolatus, Fo. vulgare,(L),(F), Casi. fistula,Che.album, Peu. graveolans	Al.sativum, L.camara,(L), (F), Cy.citratum, O. sanctum, M.arvensis		Az.indica,(D) (L),(A)
ulated with us before tment	Ad.alliacea, Al.sativum, Al.cepa, Eup.hirta, Euc-globulus, Wi.somnifera, Cl.procera, O.sanctum, Cli.lanceolatus, Fo. vulgare, (L),(F),Casi. fistula, Che.album, Peu. graveolans	L.camara,(L),(F), So. xanthocarpum, Ar.mexi-cana, M.arvensis		Az.indica,(D) (A), Cy.citra
ulated with us and insect nts) after tment	Az.indica,(D),(L),(A),Ad. alliacea, Al.sativum, Al.cepa, Eup. hirta, Wi. somnifera, So.xanthocarpum, Ar.mexicana,Cl.procera, Cli.lanceolatus, Fo. vulgare,(L),(F),Casi. fistula, Che.album, Peu. graveolans	L.camara,(L),(F),Euc. globulus, Wi.somnifera, Cy.citratum, O.sanctum		L.camara,(L) Euc.globulus, citratum, O. sanctum, M. arvensis
ulated with us and ct fed(mts) re treatment	Az.indica.(D),(L),(A),Al. sativum, Al.cepa,Eup. hirta, Wi.somnifera, So. xanthocarpum, Ar.mexicana, Cy.citratum, Cl.procera, O.sanctum, Cli.lanceolatus, Fo.vulgare,(L),(F), Casi. fistula, Che.album, Peu. graveolans	L.camara,(L),(F), Euc. globulus, M.arvensis		L.camara,(L), M.arvensis
ct fed e		Ad.alliacea,O.sanctum, Cli.lanceolatus, Peu. graveolans		Az.indica(D), (A), Al.sativ Al. cepa, L. camara,(L).(Euc. globulus Eup.hirta, Wi somnifera, So xanthocarpum, mexicana, Cy. citratum, Cl. procera, Fo. vulgare,(L),(M.arvensis, C fistula, Che. album

•Cassia, So= Solanum, Ar= Argemone, Che= Chenopodium, Az- Azadirachta, Cy= Cymbopogon, Allium, Cli= Callistemon , O= Ocimum, Ad= Adenocalymna, M= Mentha, Cl= Calotropis, Withania, Euc= Eucalyptus, Eup= Euphorbia, L= Lantana, Fo= Foeniculum, Peu= Peucedenum

Time (days) required for (++) rotting Letters in parenthesis

atments	Conc.	Time required days for development of (++) type reaction in different concen Az.indica 1	2	3	Ad.alliacea	Al.sativum	Al.cepa	L.camara 4	5	Eup.hirta	Euc.globulus
noculated and ed	S	* -	-	-	10	10	7	>15	>15	10	7
	1.0				10	10	7	>15	>15	5	7
	0.1				10	10	7	>15	>15	5	7
culated with gus after atment	S	10	-	-	7	10	10	10	10	5	10
	1.0				7	10	7	10	10	5	5
	0.1				7	5	5	10	10	5	5
culated with gus before atment	S	7	-	15	7	7	10	10	10	5	7
	1.0				7	5	7	10	10	5	7
	0.1				7	5	5	7	10	5	5
culated with gus and insect (mts) after atment											
10	S	10	10	7	-	10	7	>15	>15	10	15
	1.0				7	10	7	10	10	7	7
	0.1				7	7	5	7	7	7	7
20	S	7	7	7	7	10	7	10	10	10	10
	1.0				7	7	5	10	10	7	7
	0.1				7	7	5	7	7	7	7
30	S	7	7	7	7	10	7	>15	10	10	10
	1.0				7	7	5	15	10	7	10
	0.1				7	7	5	10	7	7	7
culated with gus and insect (mts) before atment											
10	S	7	7	7	7	7	7	15	10	7	7
	1.0				5	5	5	7	10	5	7
	0.1				5	5	5	7	10	5	5
20	S	7	7	7	7	5	7	15	10	7	10
	1.0				5	5	5	7	10	5	7
	0.1				5	5	5	7	10	5	5
30	S	7	7	7	7	5	7	10	10	-	10
	1.0				5	5	5	7	7	5	7
	0.1				5	5	5	7	5	5	5
ect fed alone (mts)											
10	S	15	15	-	-	-	-	>15	>15	15	15
	1.0				10	-	-	>15	>15	10	15
	0.1				10	-	-	>15	>15	10	15
20	S	15	15	-	10	-	-	>15	>15	15	15
	1.0				10	15	-	>15	>15	10	15
	0.1				7	15	-	>15	>15	10	10
30	S	15	15	-	10	-	-	>15	>15	-	-
	1.0				10	15	-	>15	>15	10	-
	0.1				7	15	15	>15	>15	7	10

**Az= Azadirachta. Ad= Adenocalymna. Al= Allium. L= Lantana. Eup= Euphorbia. Euc= Eucalyptus. Wi= Withania

* Only one concentration was tested.

∠ 1 - Dry powder
∠ 2 - Extract
∠ 3 - Alcohol
∠ 4 - Leaf
∠ 5 - Flower

Each value is mean of 3 replicates.

ents							latus	6	7
ulated and	S	-	10	15	-	-	10	10	10
	1.0	10	10	15	7	10	10	7	10
	0.1	10	7	10	7	10	10	7	10
ated with after ent	S	10	7	15	7	10	7	7	7
	1.0	7	7	10	5	10	7	7	7
	0.1	7	7	7	5	10	7	5	7
ated with before ant	S	15	10	15	7	5	7	7	7
	1.0	10	10	15	5	5	7	7	7
	0.1	7	7	10	5	5	7	5	5
ated with and insect s) after ent									
10	S	7	10	7	10	-	7	10	-
	1.0	7	7	7	7	15	7	7	7
	0.1	7	7	7	7	7	7	7	7
20	S	7	7	10	10	10	7	10	10
	1.0	7	7	10	10	10	5	7	7
	0.1	5	7	10	7	7	5	7	7
30	S	10	10	15	10	10	7	10	7
	1.0	7	7	10	7	10	5	7	5
	0.1	5	7	10	7	7	5	7	5
ated with and insect s) before ent									
10	S	7	7	7	7	10	7	7	7
	1.0	5	5	5	5	7	5	5	5
	0.1	5	5	5	5	7	5	5	5
20	S	7	7	10	7	10	7	7	7
	1.0	5	5	10	5	7	5	5	5
	0.1	5	5	7	5	7	5	5	5
30	S	-	7	7	7	-	7	7	7
	1.0	5	7	7	5	7	5	5	5
	0.1	5	5	5	5	7	5	5	5
fed alone (nts)									
10	S	-	-	-	-	-	-	-	15
	1.0	15	-	-	-	-	-	-	10
	0.1	15	10	-	-	10	10	-	7
20	S	15	15	-	15	-	-	-	-
	1.0	15	15	-	15	-	-	-	10
	0.1	10	7	-	10	10	-	7	7
30	S	-	15	15	10	-	-	-	-
	1.0	-	15	10	7	-	-	-	-
	0.1	-	7	7	7	10	10	-	-

**So= Solanum, Ar= Argemone, Cy= Cymbopogon, Cl= Calotropis, O= Ocimum, Cli= Callistemon
Fo= Foeniculum, M= Mentha

∠ 6 - Leaf

∠ 7 - Flower

Each value is mean of 3 replicates

Treatments		**Casi.fistula	Che.album	Peu.graveolans
Uninoculated and unfed	S	10	10	-
	1.0	7	10	7
	0.1	7	10	7
Inoculated with fungus after treatment	S	7	7	7
	1.0	7	7	5
	0.1	5	7	5
Inoculated with fungus before treatment	S	7	7	5
	1.0	7	7	5
	0.1	5	7	5
Inoculated with fungus and insect fed(mts) after treatment				
10	S	7	-	-
	1.0	7	7	7
	0.1	7	7	5
20	S	7	7	10
	1.0	7	7	7
	0.1	7	7	5
30	S	7	7	7
	1.0	5	7	7
	0.1	5	7	5
Inoculated with fungus and insect fed(mts) before treatment				
10	S	7	10	7
	1.0	5	5	5
	0.1	5	5	5
20	S	7	7	7
	1.0	5	5	5
	0.1	5	5	5
30	S	5	7	7
	1.0	5	7	5
	0.1	5	5	5
Insect fed alone (mts)				
10	S	-	-	10
	1.0	-	-	10
	0.1	-	15	10
20	S	-	-	10
	1.0	15	-	10
	0.1	15	15	10
30	S	-	-	-
	1.0	-	15	15
	0.1	10	15	10

**Casi= Cassia. Che= Chenopodium, Peu= Peucedenum

Each value is mean of 3 replicates

Table:3qFruit rot development after 15 days when tomato fruits were inoculated with *Aspergillus niger* in presence of insect, *Drosophila buscki*

Name of Plant	Intensity of fruit rot when treated with extract		
	Fungus (after treatment)	Insect (30 mts)	Insect+fungus (30 mts)(after treatment)
Argemone mexicana	3.1 a	2.2 a	3.1 a
Cassia fistula	3.1 a	1.2 b	3.2 a
Solanum xanthocarpum	3.2 a	1.1 b	2.2 b
Withania somnifera	2.2 b	0	3.1 a
Lantana camara	2.1 b	0	3.1 a
Callistemon lanceolatus	2.2 b	1.2 b	2.2 b
Eucalyptus globulus	2.1 b	0	3.1 a
Mentha arvensis	1.3 c	1.2 b	2.1 b
Ocimum sanctum	2.2 b	0	3.1 a
Calotropis procera	3.2 a	3.1 c	3.2 b
Adenocalymna alliacea	3.1 a	3.1 c	3.1 b
Azadirachta indica	2.2 b	2.1 a	3.2 b
Euphorbia hirta	3.1 a	0	3.1 b
Chenopodium album	3.1 a	0	3.1 b
Peucedenum graveolans	3.1 a	0	3.2 b
Foeniculum vulgare	3.1 a	0	2.3 a
Allium cepa	3.2 a	0	3.1 b
Allium sativum	3.2 a	1.2 b	3.2 b
Cymbopogon citratum	2.1 b	2.1 a	3.2 b

(0) Nil = No infection, (1) Poor = 25% fruit surface infected,

(2) Moderate = 25-50% fruit surface infected and

(3) (4) Severe = >50% fruit surface infected.

Means in the same column followed by different letters are significantly different at P = 0.05.

Chenopodium album for fungus inoculation after treatment; Allium sativum, Lantana camara, Cymbopogon citratum, Ocimum sanctum and Mentha arvensis for fungus inoculation before treatment; Lantana camara, Solanum xanthocarpum, Argemone mexicana and Mentha arvensis for fungus inoculation before treatment; L. camara, Eucalyptus globulus, Withania somnifera, Cymbopogon citratum and Ocimum sanctum for inoculation with fungus in the presence of insect after treatment; L. camara, Eucalyptus globulus, Mentha arvensis for fungus inoculation in the presence of insect before treatment and Adenocalymna alliacea, O. sanctum, Callistemon lanceolatus, Peucedenum graveolans for insect fed only. However, the leaf extracts of F. vulgare, L. camara have been more effective than flower extract which indicate that either the concentration of active principle in leaves is higher than flower or probably the nature of active principles in the two parts is different. In amongst the two species of Allium viz., A. cepa and A. sativum the former is more effective. Allium spp. are known for production of alliin (Stoll and Seebeck, 1951). They suggested that alliin, s-allyl-L-cysteine-sulphoxide, is converted to allicin (Diallyl-disulphide-oxide) by the enzyme alliin-lyase within a few minutes of maceration. The allicin is immediately converted to diallyl-disulphide through less known mechanism. Allicin has been reported to have antibiotic-both anti-bacterial and antifungal properties (Cavallito and Bailey, 1944; Dubrin and Uchytil, 1971). Higher efficacy of A. cepa

appears to be partly due to the presence of certain phenolic compounds such as protocatechuic acid and catechol in addition to alliin (Walker and Stahmann, 1955). The phenols in general and these in particular are already known to have antifungal properties (Pridham, 1960; Mukherjee and Kundu, 1973).

Out of the two latex bearing plants tried for the efficacy of latex, Calotropis procera has been more effective than Euphorbia hirta. The latex is known to have hydrocarbon like compounds which might probably be responsible for delaying the onset (Doby, 1965). Probably the concentration of these compounds is high in C. procera latex which is responsible for more delaying of rotting (Bonner, 1950).

Thus Lantana camara, Eucalyptus globulus, Allium sativum, A. cepa, Azadirachta indica, Ocimum sanctum and Mentha arvensis are amongst those plants which could provide protection of fruits for longer duration against A. niger and the insect.

Some of the plants tested in these studies have been found to contain substances which happen to have antimicrobial properties. Eucalyptus globulus, Azadirachta indica, Ocimum sanctum, Lantana camara and Cybopogon citratum have either volatile or non volatile substances known for supressing microbial activity (Mahadevan, 1982). Azadirachta indica in the form of dry powdered leaves or extracts has long been known as insect repellent or has been used to save commodities

against insect (Joshi, 1984; Redfern et al., 1984). Similarly, Lantana camara has been known for insect repellent properties (Attri and Singh, 1978; Pandey et al., 1979). The possibility of these substances acting against A. niger and the insect is, therefore, very great. Volatile oils of the several plants studied here, such as lemon grass Eucalyptus, mint (Morel and Rochaix, 1921) citronellol (Mahadevan, 1982) fennel (Capek, 1956), basil (Dorgich, 1971) have earlier been found effective against various microorganisms.

The results with respect to Mentha arvensis are thus in conformity with those of Sanyal and Verma (1969), and to Ocimum basilicum with Afifi (1975) against Aspergillus niger. Some of these oils have also been used for repelling insect e.g. the oil of citronella is used for making mosquito repellent cream (Metcalf and Flint, 1973).

Throughout the studies, treatment of fruits with plant extract before inoculation with fungus has been more effective than treatment after inoculation. Probably as the plant extracts penetrate the fruit either through wounds made during inoculation or other wounds made during handling provide some kind of resistance to the invading pathogens. Although many plants tested in these studies have been known for having antimicrobial properties but a very few of these have been known earlier effective against insect. Thus these plants can also be recommended for protection against fungi and insects as well.

When fruits obtained from plants grown in soil amended with oil cakes have been inoculated with fungus both in the presence or absence of insect, the fruits remained relatively free of infection for more or less 15 days except in few cases implying thereby that oil cakes provide protection against insect and fungus. Plants dipped in oil cake extracts have been found to remain relatively free of infection with nematode (Hasan, 1977). It has been suggested that during disintegration of oil cakes in soil phenolic compounds are liberated which when absorbed by the roots impart resistance (Hasan, 1977). It is likely that same mechanism might be operating here.

Normally insects have been known for transmitting the pathogens and providing wounds for enterance (Carter, 1962). In the present studies, D. busckii has not only been found to transmit the pathogen and to facilitate entrance of A. niger in tomato fruits but also aggravates the rotting and early onset of the fruit rotting. In addition to these, the insect feeding brings about changes in amino acid and ascorbic acid content of fruits. It is well known that inoculation with fungi brings about changes in amino acid and ascorbic acid content (Wood, 1967; Bhargava, and Arya, 1983) but results in changes resulting from feeding with D. busckii alone and together with inoculation with A. niger appear to be new. Moreover, treatment with phyto-extracts for controlling fruit rot both in the presence or absence of D. busckii opens a new field or fruit rot control, which is less hazardous.

CHAPTER VI

SUMMARY

1. During survey of insects in the shops of Aligarh market during the three seasons viz., rainy, summer and winter Drosophila melanogaster, D. busckii, Ephestia cautella, Liposcelis divinatoria were encountered, but the frequency of D. busckii was the highest. Rainy season appeared to favour the population of the insect.

2. In all thirteen fungi were found associated with rotting of tomato fruits, but the highest frequency was that of Aspergillus niger. The strain of the fungus isolated from the rotted fruits was pathogenic in laboratory. Most of the fungi recovered appeared to have been contracted during storage and transit.

3. The inoculum of the fungus A. niger was found to be carried through all the parts of the insect body.

4. The rotting was highest at 30°C and at R.H. 95-100 percent when not only the rotting was high but there was early onset of rotting.

5. In both inoculation of tomato fruits with A. niger and feeding with the insect separately, there was decrease in the ascorbic acid content but this decrease increased when the two were present together.

6. Feeding with insect and inoculation with the fungus brought about changes in the amino acid content of fruits. Arginine was detected in both healthy and those fed with insect; isoleucine, alanine, valine and asparagine in fruits fed by insect and inoculated with fungus. Histidine, tyrosine, lysine, cysteine and isoleucine were present in those inoculated with fungus alone.

7. Extracts of ninteen plant species belonging to eighteen genera were tried against the fruit rot caused by A.niger both in the presence and absence of D. buscki. Ethanol was effective in enhancing the time required for the development of (++) type of reaction. Ethanolic extract of Lantana camara, Mentha arvensis and Ocimum sanctum were also effective in controlling.

8. When leaf powder, water and ethanolic extract of leaves of Azadirachta indica were compared, the water extract was more effective. Similar results were obtained with water extract of L. camara, M. arvensis and O. sanctum. Extracts of A. indica, L. camara and Cymbopogon citratum were highly effective in delaying the onset of fruit rot in the absence or presence of insect. The extracts of the remaining plants were, however, effective for one or more treatments but not to all.

9. Leaf extracts of L. camara and Foeniculum vulgare were more effective than flower extracts.

10. Amongst the two species of Allium i.e., A. cepa and A.sativum, the extract of the former was more effective.

11. The latex of Calotropis procera gave better results in the control of fruit rot and insect than the latex of E. hirta.

12. Fruits from the plants grown in soil amended with different oil cakes were relatively less susceptible for attack by A. niger and the insect than those from plants grown in unamended soil.

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* Originals not seen.